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THE MEDICAL TECHNOLOGIST LOOKS AT TUBERCULOSIS*

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OBJECT—The purpose of this paper is to fulfill the need for an up-to-date and practical picture of procedures used in the laboratory for the diagnosis of tuberculosis—the lack of which has been noted in the recent literature that is readily available to the medical technologist. No mention will be made concerning the history of the disease, nor a review of the literature on the subject of it. Whenever possible, formulae will be given direct rather than just the references to it, in an attempt to make this paper a useful one.

MATERIALS USED—The following are the criteria for a good container for the collection of sputum. It must be easily handled by both patient and technician, sturdy in quality and economical in price, can be used not only in the collecting of the specimen by the patient, but also in the digestion of it by the laboratory, and can be emptied and cleaned with minimum brushing and rinsing.

An amber, four ounce square bottle with an unlined screw cap answers the above description. Heavy, blunt or pointed 50 cc. uncalibrated centrifuge tubes with corks are used for the sedimentation process. The suggestion is made to use non-calibrated tubes due to their low cost in comparison with the calibrated ones.

In the past a #14 rubber Levine tube has been used to obtain a gastric specimen, but on repeated boiling and sterilizing the tubing develops a most unpleasant odor and taste which cannot be removed. Eventually the tube becomes soft and difficult to insert. Recently, the Baxter Laboratories of Morton Grove, Ill.,

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have developed an Expendable Levine Duodenal Tube, Plastic, French size 14, 50 inches long, which is clean and ready to use. It is inexpensive and can be destroyed following its use. It is quite a relief to the patient to know that no other person has used it before him; it is tasteless and a little stiffer, so that it is not as easily coughed up as the rubber ones. The use of a sterile mineral oil as a lubrication is a matter of choice, but putting the tube on ice is unnecessary.

The gastric tray which is approximately 3" x 6" x 9" is equipped with several four-inch gauze squares, a four ounce amber square bottle with metal cap, not screwed on tightly and a 20 or 30 cc. syringe. It is wrapped with two towels, autoclaved at 20 pounds for 15 minutes. The inside wrapper is placed on the patient's lap, care being taken to keep the tray as clean as possible. MEDIA—Various types of media have been used in the past by this laboratory, but Petragnani's is still preferred for the original isolation of the tubercle bacillus. Some technologists make their own, but we purchase ours at the Difco Laboratories in Detroit or the Albimi Laboratories in Brooklyn, New York. The choice really depends upon the conditions at the laboratory, and the time and aptitude for making the media. For those who have the ability to do so, the following is the formula.*

Milk	900 cc.	Peptone	6 gm.
Potato flour	36 gm.	Eggs	24
Egg yolks	6	Glycerin	72 cc.
Pieces of potato	6 Size of an egg		
2% aqueous solution of malachite green		60 cc.	

Cut the potatoes in thin slices, add the milk, potato flour and peptone, and cook in a double-boiler for two hours, stirring constantly until sticky, then stir occasionally. Break eggs and egg yolks into a two liter flask and shake well. Add glycerin and malacite-green and shake. Cool the potato mixture to 45-50° C., add the glycerin-dye mixture, shake, filter through gauze, dispense in tubes and place on tray in a slanting position. Inspissate according to instructions in Todd and Sandford, tenth edition, page 773.

In comparing Petragnani's media with Petroff's, it was found that the cultures were very slow in growing on the latter and some did not grow at all. The American Trudeau Society has developed a media, called the A.T.S. Media, which can be obtained at Difco. It gives comparable results with that of the Petragnani's. The Difco Bovine culture media for the isolating of the bovine type of Mycobacterium tuberculosis is the only known commercial Bovine Media available at this time. A word of advice, keep ahead on your supply of media when it is pur-

*A.P.H.A., Standards and Reagents.

chased from any laboratory supply house, as it often takes a long time for an order to be filled.

STAINS AND REAGENTS—A 4% NaOh solution and a 3.6% HCl solution are used in the digestion and concentration of material for acid-fast examination. When large quantities are used, it is a timesaver to make up the solution in five liter bottles, and transfer them to pouring flasks for daily use. The formulae are as follows:

4% NaOh;	160 gm. NaOh pellets 4,000 cc. distilled water
3.6% HCl;	108 cc. concentrated Hydrochloric acid 3,000 cc. distilled water

The staining procedure used is one of the many modifications of Ziehl-Neelson, but it is quick and reliable, and many racks can be done in a relatively short time. The carbol-fuchsin is made in a stock solution and diluted as required.

Stock Carbol-Fuchsin;	1 gm. Basic Fuchsin 5 cc. Phenol 5 cc. Glycerin 10 cc. 95% Alcohol
Diluted, working solution;	20 cc. stock solution 280 cc. distilled water

The working solution is kept in a closed copper container, 1½″ x 3″ x 7″, and placed over direct heat when staining is to be done. The decolorizing agent and counterstain are kept in covered glass containers, approximately 4½″ x 3½″ x 8″ in size. The 3% acid alcohol is made in the laboratory using hydrochloric acid, but the Alkaline Methylene Blue is purchased from a laboratory supply house.

A 5% solution of Old Tuberculin is purchased from Parke Davis or Lederle's, and is prepared aseptically and used for the skin testing of the guinea pigs, when they are used for identifying and isolating the bacilli.

CARE OF EQUIPMENT—All glassware containing the residue of any specimen is autoclaved at 20 pounds for one hour. The equipment is then rinsed and brushed until clean and then immersed into a potassium-dichromate sulfuric acid cleaning solution over night. It is then rinsed again in tap water and washed in a solution of trisodium phosphate, rinsed both in tap and then distilled water and placed in an oven for drying. All bottles and centrifuge tubes are capped or corked and dry-air sterilized for one hour at 150° F., before use. After so long a time, the bottles become stained and discolored. Prior to delivery to a patient, a label is placed on the bottle beside the identification label, which reads, THIS BOTTLE IS BACTERI-

*E. R. Squibb and Son.

OLOGICALLY CLEAN. Rubber goods, syringes and needles are placed in a 50% phenolor* solution for three hours, rinsed, washed and rinsed again in tap and distilled water, air-dried, wrapped and autoclaved. About every four months, sterility tests are run on all equipment, reagents and media for any contamination by the tubercle bacilli. To date all cultures have been negative.

PRECAUTIONS—Many have asked and will continue to do so, "Aren't you afraid of getting tuberculosis, working in such a hospital?" Our answer is "No," for these reasons. We know what we are dealing with, respect its potentialities, and take proper precautions. At the time of employment, medical, technical, nursing and lay personnel are oriented individually, as to the care and precaution he must take. That is to the proper way of putting on a mask and gown and then removing it; the right way to wash and rinse hands, the use of clean and dirty areas. In the laboratory, every specimen and piece of equipment is considered contaminated, that is from the urine bottle on the table to the small adjustment of the microscope or the pencils and record books. When handling contaminated material and the animals, or contacting the patients in anyway, the technicians and animal caretaker wear masks and gowns, and gloves if there are any skin abrasions on the hands.

Unprotected ultra-violet lights are hung low from the ceiling over the working counters, and are kept on at all times when the Staff is out of the laboratory. The lights are inexpensive, made by Westinghouse, called Sterilamp, and do a great deal in cutting contamination both in the laboratory and the guinea pig room. The rays do not penetrate glassware, therefore are not harmful to any reagents or living cultures. Guinea pigs are apt to get sun-burned eyes if kept too close to the lights in their separate room, but placing the cages at a distance of six feet or more from the lights will prevent this from occurring.

Phenolor is used very liberally. The counters are washed once a day with the straight solution and a sponge. Caution—Use rubber gloves, it is hard on the hands. Most unpleasant odors can occur on concentrating or autoclaving specimens. This is remedied by using a deodorizing bomb, which should be on hand at all times. There are many on the market, of all kinds and prices. There are two MUSTS to consider, that they are Non-toxic and Non-inflammable.

Common sense is used by the technologist in the planning of her work; that is she doesn't run a series of chemistries at the same time she is concentrating sputums. No pipette is ever used to measure a solution and then returned to the bottle for use at a later time, but put aside as used and a clean one obtained from a closed container when required. A Dutch oven about

18″ x 21″ x 13″ in size was purchased at a hardware store for the drying of slides. Legs were put on it of the required height of seven inches, so that a Bunsen Burner could fit under it, and a metal bottom inserted in the oven on which to put the slides. In this manner, a precaution is taken against a sudden gust of wind which might blow off a tiny droplet from the slide full of acid-fast organisms. It should be remembered that there are people in and out of the laboratory some of the time, who might not have on a mask. Every member of the hospital staff has a periodic x-ray, those in the laboratory have one every two months.

This all may sound like a terrific ordeal, "Precaution," but on analysis it's all a matter of forming a few good habits which are fairly sensible and easily carried out. No exact figures could be obtained from the United States Public Health Service as to the number of laboratory personnel who have contracted tuberculosis, but we all know of others in different branches of the medical service who have done so.

COLLECTION OF SPECIMENS—Every laboratory has its own "pet" system for collecting specimens. This one, whose aim primarily, is the isolation of the tubercle bacillus, has such a set routine. Upon admission, the patient is given a one ounce corked bottle and instructed to place his early morning sputum in it by coughing from the bronchi and lungs. Impress upon him the inadequacy of a little saliva. The specimen is digested and concentrated, smear and stained, then, examined for the presence of the acid-fast organism. If none are found, or just a very few that are recorded as a Gaffky I, the patient receives a four ounce amber bottle, with the same instructions for collecting his sputum for five consecutive mornings. This material is treated as the one specimen, and if found negative, or a Gaffky I or II, the sediment is cultured.

The mechanics of inserting a gastric tube will not be discussed in this paper, but it should be realized by the technologist, that it is very natural for any patient to be apprehensive of the prospect of a gastric lavage. Our procedure is to visit the patient the day before his first test is to be done. A simple explanation is given as to how the tube is inserted, there being no pain, but just slight discomfort. Make him realize that the success of the test depends a great deal upon his cooperation. Show him the tube, allow him to feel it, and if the occasion seems apropos, go into a short discussion as to what tests are run on the material collected. Instruct him and the nurse that no fluids are to be taken after midnight, and breakfast should be delayed until after the test is completed. Here again the technologist should remember her precautionary measures. An out-patient may come into the laboratory for a gastric with orders for a chemical analysis. He also, may have tuberculosis. If necessary, you can

develop a hypothetical cold, but whatever the ruse, WEAR A MASK.

At the time of the test the technologist must be prepared for all types of reactions of the patient. He may swallow the tube, not cough or gag, and cooperate 100%. Most of them do after the first test. Others may be frightened. At this time speed and confidence in your manner, a quiet tone of voice, reassuring him that he is doing fine is most helpful. Some patients may be just belligerent, due chiefly to fear, saying they can't swallow the tube any farther, etc. Here speed of action and often a gentle scolding is required. It should be remembered that material must be collected as quickly as possible. To put it off until another time only prolongs the suspense of the patient.

Transudates and exudates are usually collected by the nurses or doctors and sent to the laboratory. If the opportunity arises to specify the type of specimen you will require, suggest that the entire amount collected be given you. Whenever possible urine for the culturing and animal inoculation should be obtained by catherization. There are times when material to be examined can only be obtained by swabbing the area. If the doctors realize that just a small amount of material is more usable in a syringe than on the swab, they will do their best to get it in that manner.

PREPARATION OF THE SPECIMENS—It should not be necessary to mention this but the fact should always remain in the upper-most thoughts of the technologist's mind, that care must be taken to prevent a mixup of any specimens. Here a routine must be set up and adhered to firmly. It should be simple, but fool-proof; such as, a dozen specimens are to be done. Line them up alphabetically, number consecutively, and enter the entire information as to name, number and so on, on whatever type of report blank is used. Carry the numbers through on the tubes, slides and culture tube-labels, which should also have the complete information. When making any type of transfer, be sure that the numbers on each container are checked.

Sputums, Gastrics, and Bronchoscopies

Transfer each specimen to an individual, four ounce, square amber bottle. The amount of 4% NaOh added to each depends upon the consistency of the specimen. If very mucoid and heavy, add an equal portion of it: if the specimen is thinner, with only a small number of mucoid bits, as a gastric or bronchoscopy specimen often are, add just a small amount of the digesting material. If the bottle is more than 1/3 full, divide it. Place the bottles tightly capped in the Kahn shaker and shake for five minutes. Then examine for an even suspension. Some of the heavier specimens may require a few more minutes of shaking.

If your shaking machine is old and noisy, ask your maintenance man to make an insulated box with a tightly fitting cover and screw it onto the table over the machine. This will do a great deal toward cutting down on the noise.

Pour about 10cc. of the digested specimen into a 50cc. centrifuge tube, which has been checked for cracks, matching the numbers on the bottle and the tube. Save the bottle until the test is finished, for tubes do sometimes break in the centrifuge. Add sufficient amount of 3.6% of HCl, drop by drop until a flocculation is formed throughout the suspension. The tubes are recorked, balanced on a scale and centrifuged at 4,000 R.P.M. for five minutes. A balancing fluid of sterile saline may be used. Pour off the supernatant fluid into an enamel pitcher containing 15-22 cc. phenolor, recorking and wiping the drip from the side of the tube with a gauze square.

Another procedure of digestion and concentration for sputum or gastrics very frequently used is that of adding an equal amount of Hank's solution to the material in a 50cc. centrifuge tube. Shake thoroughly and digest at 37° C. for 30 minutes. Neutralize with 12N HCl to a pH of 7.0, or until a precipitate appears and centrifuge at 2,000 R.P.M. for 10 minutes. Decant the supernatant fluid, smear and stain the sediment. The formula for Hank's solution is as follows:

NaOh pellets	40 gms.
Potassium alum	2 gms.
Brom Thymol Blue	.02 gm.
Distilled water to one liter	
Autoclave 15 lbs. for 20 minutes	

There are many other procedures for the digesting and concentrating the sputum, gastric and bronchoscopy specimens. It must be remembered that a few do not permit the use of culture or animal inoculation after the digesting procedure.

Spinal Fluid—Whenever possible request the spinal fluid to be collected in two separate tubes. Place one on the counter for 24 hours for pellicle formation, this is not often seen in our experience. The other can be used for routine bacteriology, cell count and chemistry. At the end of the 24 hour period, examine the specimen for a pellicle. If present, divide it with a pair of sterile loops, placing a part of it on a sterile slide and the other on a culture slant. If no pellicle has appeared, add one to two drops of 5% tannic acid, and centrifuge. Or, add sufficient amount of 95% alcohol, drop by drop until the suspension is milky and centrifuge. Decant the supernatant fluid, smear and stain the sediment. It has been noticed that in the use of tannic acid, if the sediment is allowed to stand awhile before culturing, the

slant will turn black when inoculated, but this will not hinder the growth of the organism.

Miscellany—Check the pH* of urine and clear pleural fluid and adjust to pH 6.5, centrifuge and decant the supernatant fluid. Any thick pleural, abdominal or joint fluid is digested and concentrated as sputums are. Gastrics are not smeared. Often times the hay or butter bacillus found on leafy vegetables or fruits survive digestion and concentration, and can be confused with the pathogenic organism when seen on smear. The diagnosis there is made from the results obtained from the culture and animal inoculation. Emulsify feces in saline, centrifuge, decant, smear and culture but do not inoculate into an animal. Bone marrow and blood should be obtained in sterile balanced oxalate. Lake with sterile distilled water. centrifuge, smear, culture and inoculate into an animal as required.

STAINING PROCEDURES—Using only new slides with one end frosted, number with a lead pencil according to the original specimen. Flame slides gently and with a sterile loop, cover a 2/3 portion of the slide with the prepared sediment, and place in the oven for drying. Bacteriological technique must be used throughout the whole procedure. The slides may be left in the closed oven for an hour or more to dry naturally or may be hurried with the use of a very low flame from a Bunsen Burner, taking particular care though, that the flame is not placed directly underneath the slides. When dry, each slide is flamed quickly and placed in a metal rack.

The carbol-fuchsin is brought just to boiling and then the heat removed. The rack is inserted for 5 minutes, washed carefully under a tap-water faucet, drained quickly, placed in the acid-alcohol bath for the same time, washed and drained as before and finally into the methylene blue for 3 to 5 minutes; wash, drain and air dry. Other counterstains may be used, such as picric acid, or brilliant green, all a matter of choice. When there are just 2 or 3 slides to be stained, Corper's procedure may be used. Cover the slides with carbol-fuchsin and steam gently by direct heat for 5 minutes. Allow to cool for 20 minutes. This step is important, as it allows the stain to precipitate into the organisms. Decolorize with acid alcohol until a faint pink, wash with tap water and counterstain with methylene blue, wash, dry and examine. When any staining procedure is not used at frequent intervals a known positive should be run along with the unknowns to check the reliability of the stain.

EXAMINATION OF THE SLIDES—The slides are examined by oil immersion, covering the whole smear before it is reported out as negative. The positives are reported according to the

*Nitrazinc Paper—E. R. Squibb and Son, Cleveland, Ohio.

number of organisms seen, an average per field, using the Gaffky Scale;

Gaffky	I	1-4 bacilli per slide
Gaffky	II	1 bacilli on average of several fields
Gaffky	III	1 bacilli on average of each field
Gaffky	IV	2-3 bacilli on average of each field
Gaffky	V	4-6 bacilli on average of each field
Gaffky	VI	7-10 bacilli on average of each field
Gaffky	VII	fairly numerous average on each field
Gaffky	VIII	numerous average on each field
Gaffky	IX	very numerous average on each field
Gaffky	X	numerous masses on each field

CULTURE TECHNIQUE AND THE EXAMINATION OF CULTURES — Using bacteriological technique, sufficient amount of the sediment is poured onto a slant of Petragnani's media, any excess being returned to the original tube. Prior to the inoculation paste upon the culture tube a label, having good adhering qualities, with complete information written on it as to the patient's full name, the date, type of specimen and so on. Incubate the tube horizontally at 37° C. Wash the sediment with sterile saline, centrifuge and decant three times, for inoculation into guinea pigs.

The cultures are examined once a week macroscopically for the presence of the "typical" colony, which may be a pale yellow and rough or a darker color, almost to an orange and shiny. The negative tubes are reincubated for 8 weeks to 2 months before reporting out as negative. Over 1,000 negative (macroscopically) cultures were smeared and stained to determine if there were organisms growing, but could not be seen with the naked eye. Only 1 culture showed organisms by smear, at the end of 2 months. Upon reincubation for another 2 months, no more could be found on the slant, by smear and therefore was reported out as negative. The colonies from the positive slants are smeared and stained and examined for the acid-fast qualities and reported as "culture of such and such material as positive for acid-fast organism" on the date isolated.

No hard and fast rule has been established in routine laboratory technique for the determination of pathogenic organisms, other than culture and animal inoculation. If there is such a question the organisms taken from the culture are emulsified in sterile normal saline, 0.1cc of which is inoculated into a guinea pig. Studies have been made in the laboratory, comparing the color and character of the colony with the microscopic findings, that is in the arrangement, size, presence and absence of Much's granules. No definite conclusions have been reached. At the present time further studies are being run in an effort to determine by a simpler method the pathogenicity of the organism.

It is most time consuming and expensive to inoculate every positive culture into an animal.

There is a difference of opinion among many as to the necessity of both animal and culture inoculations. Realizing that some workers have estimated that 100,000 organisms/cc. of concentrated fluid are required for them to be seen on a smear, we feel that a series of negative smears are of no value and that even several negative concentrated smears should not be relied upon for a final diagnosis.

It is the policy of this laboratory that all negative concentrated sputum smears, other than admission specimens are cultured, all gastrics are cultured and inoculated into animals, and all transudates, whether the smears are positive or negative, are cultured, and inoculated into animals when required.

It might be of interest to note a comparison of results between cultures and animal inoculation, in relation to the type of specimen. Of the 120 gastric lavages that have recently been obtained,

3 cultures, negative, with positive pigs
27 cultures, positive, with negative pigs
6 cultures, positive, with positive pigs
84 cultures, negative, with negative pigs

A total of 20 sputums showed,

1 culture, positive, with negative pig
2 cultures, positive, with positive pig
17 cultures, negative, with negative pig

There is a closer parallel between the 138 bronchoscopies,

12 cultures, positive, with negative pigs
17 cultures, negative, with positive pigs
21 cultures, positive, with positive pigs
88 cultures, negative, with negative pigs

ANIMALS—Most hospitals do not have the equipment, space nor personnel to breed and raise their own animals. The best results are obtained by purchasing them from a local reliable animal breeder, specifying quantity and weight of each and the time of delivery. With the assurance of regularly receiving good stock, the animals can be used without the preliminary isolation period and skin testing. Animals received from an unknown breeder or those which have been shipped from a long distance must be isolated for at least 2 weeks and during that time, skin-tested for tuberculosis. Over a period of 30 months, this laboratory used both the long-distance and local sources for its supply of guinea pigs. Shipping the animals by express was often disastrous, and most time consuming. There was a matter of proper feeding and heating facilities during shipment

and often the animals sickened and died in transit or soon after they were delivered. The local supply is ideally successful. Delivery can be made on a 24 hour notice. The source is known to be reliable and is used by many other hospitals in this city.

The new animals are kept in two cages away from the inoculated ones, separated according to sexes. After the material is prepared, 0.1cc is injected into the right inguinal region of a tuberculin-anergen animal using a tuberculin syringe and a 24 or 27 gauge needle. Following the injection the pig is isolated in his own cage. Pertinent information is kept on each pig; the number of the cage, the description of the animal, the patient's name, date, and type of specimen injected. This information is most successfully kept in an animal book, together with dates for skin tests and autopsies. In this manner a great many of the tests or post-portems can be done at one sitting.

Guinea pigs, or any other animal should be kept in a room or animal house which is dry, well ventilated, with temperature control and facilities for easy cleaning of the cages and disposal of refuse. This sounds like a large order but every detail which can be carried out in the proper care of the animal produces satisfactory end results, that is the animal remains alive until his time is scheduled for sacrificing and autopsying.

Individual wire cages are used with removable refuse trays and feed pans. A layer of sawdust in the refuse trays aids in controlling the ammoniacal odor. These are flushed out with a hose down a wide-mouthed drain every other day and allowed to dry. Any food left over that is carrot tops or lettuce is removed immediately. These particles of food degenerate fast. The pig may become hungry and he will eat these remains. His inability to vomit after eating this material will give him diarrhea and in a couple of days he will die. Small metal feed pans which can be hooked onto the sides of the cages are more practical for the dry feed than a dish. The pan can be hooked up at a level which will discourage the animal from nesting in it. A round pellet of salt is hooked up high so that the pig can just reach it.

The animals are fed ⅓ to ½ a head of lettuce a day, depending upon the individual requirements of the animal. Some are big eaters and some are not. The head lettuce is cut to the required size and only the rusty portions removed. The outside green leaves are what the animals need in obtaining their Vitamin C. The absence of this vitamin lowers their resistance to bacterial infections. The Purina* is placed in the feed tray once every other day, and a good size carrot with the green tops left on are given twice a week. They make a good week-end

*Purina Chow—purchased at any feed store.

feeding with tops removed. Sufficent water is obtained from the washed lettuce. The temperature of the room should remain constant, varying 2-4° at the most, in the range of 68-72° F. A great deal depends upon the local situation. If there is any chance for a noticable variation or a sudden change in weather, frequent inspection should be made during the day and night. During the hot weather, 75-80° F. in an animal room does not seem to harm them when there is plenty of fresh air, but no drafts at any time. Changes in the weather during the spring and fall cause the most difficulty.

The personnel caring for the animals should take strict precautions for themselves and for the animals. It is well-known that the guinea pig is most susceptible to any respiratory infection. One of them can become ill with an acute infection from a contact with a worker who may not realize he is ill at all, and in a few days that infection can spread throughout your colony and a great loss is sustained. For that reason a worker always wears a mask, and gown and gloves.

When the animal is removed permanently from the cage, the salt pellets are destroyed, the tray, feeding pan and all refuse removed, and the whole cage thoroughly cleaned with hot water and phenolor. It is allowed to dry for 24 hours, before using again. Guinea pigs do not respond to any medication in the treatment for respiratory infection, other than the use of camphor vapors. Penicillin injections are fatal to these animals, nose drops cause pneumonia and death. A simple remedy can be used to restore a sick pig, by placing him in a wooden box containing an inch of sawdust, his regular feeding, and then sprinkle camphor along the sides of the box and cover it, not tightly, with a piece of cotton cloth, leaving two inches for an opening to prevent smothering. The camphor should be replaced once a day. In 2 or 3 days, the pig usually revives if he has been treated soon enough. In handling the animals, watch for a fast pulse, running eyes and extreme warmth.

Three weeks following inoculation, the guinea pigs are shaved on the belly and skin-tested, using a 0.1cc. 5% solution of O.T. from a 27 gauge needle on a tuberculin syringe. Care should be taken in holding the animal firmly, so that he will not squirm. At the end of 48 hours, the skin tests are read and reported as positive or negative. If negative, at the end of the second, three-week period, they are again skin-tested and reported accordingly. Two months from the date of inoculation, the animals are sacrificed by placing them in a tightly sealed can. Chloroform is used as the euthanasia for about 45 minutes. The animals are removed to the autopsy table and allowed to air. They should be autopsied as soon as possible, at least within two hours from the time of the sacrificing, as they are

more difficult to manage when rigor mortis has set in. In order to do a number of animals at one time, each pig is wrapped securely with a 2-inch width of gauze on which has been written in red pencil, the number of his cage. The number on the band and its color, description, etc., are compared with that in the animal record book.

The purpose of the autopsy is to remove the viscera for macro- and microscopic studies, with as little trauma to the organs as possible. Trim off the fur to the skin on a small area in the region of the lower bowel. With a small hemostat, grab the layers of skin and make a midline incision, being careful not to puncture any of the intestine. The use of a sharp, stainless steel suture scissors, both ends blunt, are the most desirable. Continue the invision to the top of the sternum. Cut away the ribs from each side of it. The sternum is now free so that the animal may be held firmly by the hemostat. Sever the diaphragm on each side to the back and loosen any adhesions. The windpipe and aesophagus are cut and any other material adhering to the spinal cord, down beyond the kidneys. By this time the entire viscera is freed and falls out. The lungs, heart, liver, spleen and kidneys are removed in their entirety and placed in a jar containing sufficient amount of 10% formalin to cover the specimen completely. The jar should be labeled with the patient's name, the type of specimen, the dates of inoculation and autopsy. The viscera should be examined grossly for any pale yellow tubercules, and recorded in the animal book as to whether any were seen or not. This bit of information is often useful to the physician, before the microscopic report is received from the pathologist. It has been found very helpful if the animal man is informed the day prior to that set for the autopsies, so that he does not feed the animals on the day of their sacrificing. Getting around a full stomach is tedious and often most unpleasant if it has been punctured. Upon receipt of the microscopic report, comparison notes are made with those of the macroscopic notations in the animal book.

STREPTOMYCIN SENSIVITY TESTS—A great many procedures for the streptomycin sensitivity determinations are still in the research stage as far as the technologist is concerned who works in the laboratory of a general hospital. The literature is voluminous and the procedures, although complicated are in great detail. There are several factors a technologist should consider when setting up such a routine—the availability of special equipment, a sterile room in which to work, and the frequency of orders for such determinations. It is most advisable to study the various aspects of the procedures. A great deal of knowledge can be obtained by referring to the authors Dubos, Williston and Youmans; references for such articles

are given at the end of this paper. Unless there is a sterile room where a technologist can work undisturbed, it is very difficult to make the special media required in the sensitivity test without contamination, because of addition of measured amounts of several sterile constituents. To date this laboratory finds the most economical in time and patience, although the initial expense is high, is to order from the Albini Laboratories in Brooklyn, New York, tubes of Dubos' media, with varying amounts of streptomycin added. It takes from 2 to 3 weeks for an order to befilled.

Procedure—On the obtaining of a good positive culture of the Mycobacterium tuberculosis, the slant of Petragnani's media is covered with a 0.01 M solution of Potassium Phosphate (0.136gr/100cc. distilled water; autoclave 20 lbs. for 15 minutes). Scrape off the colonies with a sterile loop into the solution and allow it to stand undisturbed for 30 minutes. The large pieces of media will settle to the bottom of the tube and the organisms remain in the suspension. Make a smear of the suspension and stain for the acidfast organisms, as a check for the number of organisms and any contaminants. Using a sterile tuberculin syringe with a 27 gauge needle, draw it about 2/3cc. of the suspension. With asceptic technique, add 5 drops to each of the tubes of media prepared for the test.

Incubate at 37° C. for a week and after gently shaking, examine for gross flocculation. Reincubate for another week and make a final examination, reporting the highest concentration of streptomycin where the flocculation is seen, "organism grows at such and such concentration of Streptomycin" and then note the next tube where there is no flocculation, reporting, "organism is sensitive to such and such a concentration of streptomycin." If the fluid media shows a growth that is fine, smooth and evenly distributed, another organism is present. If the control tube has no floccular growth in it, then there were not sufficient organisms in the suspension. This procedure is a combination of several and seems to work very well. It is time consuming in that a culture must be obtained before the test can be started.

COMPARISON OF HUMAN, BOVINE AND AVIAN TUBERCULOSIS—

The bovine organism is shorter and less slender than the human type but it is impossible to differentiate them on the acid-fast smear. They grow less prolific on artificial media, the growth not being enhanced by the presence of glycerin in the formula. After intravenous injections rabbits die from it in 2 to 5 weeks, while in the human type, they develop only a mild disease which persists from 2 to 6 months. Guinea pigs die from it more quickly than from the human type. Difco Bovine media

or Lowenstein's (which cannot be purchased commercially) without glycerin are both suitable for the growth of the Bovine organism. The following is the formula for Lowenstein's media:

Asparagin	3 gm.*
Potassium Phosphate	1 gm.
Magnesium Sulphate	1 gm.
Glycerin	60 cc.
Distilled water to 1 liter.	
To 600cc. of the above add	
Potato starch solution, 50%	10 cc.
Tomato juice	60 cc.
Eggs	16
Egg yolks	4
Congo Red, 2% aqueous solution	20 cc.

Dispense into tubes in 5 to 6cc. amounts, lay on trays and inspissate according to instructions given in Todd and Sandford, Tenth Edition, page 773.

The following procedure for the typing of the two organisms is recommended by Park and Krumweide and can be used in most any laboratory where rabbits are available:

1. Remove and place on sterile filter paper the growth from a tube of culture media using a platinum spatula or a coarse wire loop.
2. Weigh (must not be less than 3 to 5 mg.)
3. Transfer the mass to an agate mortar to which one drop of sterile beef bile is added and grind thoroughly.
4. Divide into 2 parts and add sterile physiological saline, so that in one sample, each ml. contains 0.1mg. of culture and in the other specimen 0.01mg. of culture. If the amount of culture is insufficent for weighing, make the suspension of the organism in the saline, transfer it to two sterile Hitchcock centrifuge tubes, centrifuge, measure the sediment and dilute according to the required strength.
5. Select two rabbits weighing 1500 to 2000gms. Inject intravenously into one rabbit 1cc. of the suspension containing 0.1mg. of the culture, and into the other rabbit 1cc. of the suspension containing 0.01mg.
6. Record weight of the rabbits at the time of injection and death; the animals surviving will be killed and examined at the end of eight weeks. The bovine type will kill the rabbit within two months. At autopsy the animal will show extensive pulmonary lesions. The human type will not kill the animal but may produce slight discrete lesions in the lung.

One case of avian tuberculosis was found at this hospital on a female patient. Repeated positive concentrates failed to grow

* General Biochemicals, Inc., Chagrin Falls, Ohio.

on any media, nor develop in guinea pigs. Two chickens were skin tested on the wattle with Avian Tuberculin, produced by the Ft. Dodge Serum Co., Ft. Dodge, Iowa, and were found to be negative. A week later the control chicken was fed with a suspension of known avian tubercle organisms, and the other fed a suspension of the unknown organisms. Four and six months later approximately, the chickens died and both were found on pathological examination to have had pulmonary tuberculosis.

SUMMARY—An attempt has been made to present a practical, over-all picture of the laboratory diagnosis of tuberculosis.

REFERENCES

1. American Public Health Assn., Volume on Standards and Reagents.
2. Bradbury, F. C. S., Human Tuberculosis Due to Avian Tubercle Bacilli. A Report of a Case. Lancet, January 19, 1946.
3. Corper, H. J., The Certified Diagnosis of Tuberculosis, Jr. A. M. A. 1928, 91. 371.
4. Baldwin, Edward A., Non-pathogenic Acid-fast Bacilli. Amer. Rev. of Tuberculosis, June, 1942. No. 6. Vol. XLV. 756.
5. Hanks, H. J., Methods of Concentration. Jr. Lab. and Clin. Med. Vol. 23, 1938. 736.
6. Kolmer, John A. and Boerner, Fred, Approved Laboratory Techniques. 4th Edition. Appleton-Century Co., Inc.
7. Lande, Frank and Dixon, R. F., A Comparative Study of Guinea Pig Inoculation and Cultures for the Demonstration of Tubercle Bacilli. Technical Bulletin of The Registry of the A. S. C. P., September, 1944, Vol. 5. 93.
8. Levinson, S. A. and MacFate, R. P., Clinical Laboratory Diagnosis, 3rd Edition. Lea Phebiger.
9. Park, W. H. and Krumweide, C., The Relative Importance of the Bovine and Human Types of Tubercle Bacillus in the Different Forms of Human Tuberculosis. Jr. Med. Research 23; 205, 1910.
10. Ritterhoff, R. J. and Hurrell, Loretta, Demonstration of Tubercle Bacilli. Survey of Methods Used in Ohio and Recommended Procedures. Ohio State Medical Journal, Vol. 42, Sept., 1946. No. 9.
11. Roberts, Arwyn, Modern Methods of Culturing M. Tuberculosis, with Special Reference to the Dubos Technique, Tubercle Vol. xxlx. May 1948, No. 5. 97.
12. Todd, J. C. and Sanford, A. H., Clinical Diagnosis. Tenth Edition. W. B. Saunders Co.
13. Topley, W. W. C. and Wilson, G. S., Principles of Bacteriology and Immunity. Second Edition. William Wood.
14. Tuberculosis Evaluation Laboratory Methods, Communicable Disease Center, U. S. P. H. S., Atlanta, Georgia.
15. University of Illinois, Dept. of Bacteriology. Mimeographed Laboratory Notes. 1941.
16. Williston, E. H. and Youmans, G. P., Streptomycin Resistant Strains of Tubercle Bacilli. Amer. Rev. of Tuberculosis Vol. LV. No. 6. June, 1947. 536.
17. Ibid, Factors Affecting the Sensivity in Vitro of Tubercle Bacilli to Streptomycin. Ibid. Vol. 59. No. 3. March, 1949.
18. Worden, A. N., The Care and Management of Laboratory Animals. Williams and Wilkins, 1947.
19. Smith, D. T. and Martin, D. S., Zinnser's Textbook of Bacteriology, 1948 Appleton Century and Crofts.

LABORATORY TESTS IN SUSPECTED VIRAL DISEASES*

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Although some of mankind's oldest ailments including smallpox, influenza and the common cold are virus diseases, scientists are woefully ignorant of many aspects of viruses. Unknown diseases of possible viral origin and "new" viruses of undetermined importance are constantly turning up. The possible role of viruses in the whole field of malignancy remains undetermined. It is not surprising that Barnard writes, "It is no exaggeration to say that virology stands today about where bacteriology stood in the early 1900's." Nevertheless, in recent years much progress has been made in the diagnosis, control and treatment of viral diseases, and virology has become almost as important as bacteriology in medicine and public health. With the knowledge that most of the rickettsial and some of the viral diseases are amenable to specific treatment, physicians increasingly are seeking laboratory information in dealing with suspected viral and rickettsial diseases. Laboratory tests which offer real help to the perplexed physician in the differential recognition of viral diseases have become increasingly more practical and widely applicable.

Since most of the procedures in virology of interest in the medical and public health laboratory have been essentially research procedures until quite recently, the opportunities for those seeking training in this field have been limited. It is lamentable that there are few well trained virologists, but it is understandable that there are so few virology laboratories. Opportunities for learning some of these procedures are now provided in the Texas State Health Department and other laboratories.

Collection of Specimens

Generally speaking, in searching for the causative virus or rickettsia the earlier the appropriate specimen is collected the greater is the possibility of a successful outcome. Since the causative agent is likely to die out otherwise, specimens should be submitted in a chilled or frozen condition. The kind of specimen desired depends upon a number of factors concerning the disease in question. If, as in the average laboratory, serological tests only are performed, nothing is more desirable than a tube of clotted blood obtained as early in the illness as possible

* Read before ASMT Convention, June, 1950, Houston, Texas.

and followed by "repeats" in a manner to permit detection of a possible rising titer as soon as possible. It is a good policy in the hospital dealing with virus diseases to draw blood specimens routinely shortly after admittance and immediately prior to release of the patient. Post mortem tissues frequently are a fruitful source of the virus and should be preserved in glycerinated saline or kept in the frozen state. When the physician is in doubt concerning the collection of specimens he should be encouraged to contact the virologist. The virologist should be advised of the disease or diseases suspected, because of the growing list of tests available.

Recovery and Identification of Viruses

The isolation and identification of viral and rickettsial agents remain largely in the realm of difficult, time consuming, expensive and frequently dangerous research procedures. I agree with Smadel[2] that the physician be advised with few expections to refrain from requesting the isolation and identification of the virus unless he is willing and able to cooperate fully in the investigation. When a serological procedure is not readily applicable or available the recovery of the virus is the logical approach to the identification of the disease.

Embryonated eggs and mice are most widely utilized for cultivation of viruses. Several viruses multiply rapidly and cause lesions on the chorio-allantoic membrane of the embryonated egg and this fact is the basis of a rather speedy presumptive test for smallpox.[3] The amniotic sac is the best tissue for recovery of some viruses, including the mumps and influenza viruses. The yolk sac is favored for cultivation of rickettsia, as was first shown by Cox,[4] and viruses of the psittacosis-lymphogranuloma group of viruses. The allantoic fluid is also a favorite inoculation site in embyronated eggs, and is widely utilized in the cultivation of viruses for antigen production.

White mice are widely utilized because of the great variety of viruses to which this animal is susceptible. Suspectibility to laboratory infection is somewhat dependent upon several factors. The intracerebral route of inoculation is widely used and especially with suspected neurotropic infections. In suspected Q fever and psittacosis, the intraperitoneal route is perhaps preferable. "Blind passages" are sometimes employed to render the infection more virulent or at least more apparent. Infant mice are utilized for recovery of the Coxsackie virus. On the other hand, young mice appear to be less susceptible to the Lansing type of poliomyelitis.

When the virus is recovered its identity must be established. Identification of an unknown virus frequently depends largely upon determination of host or tissue susceptibility range and

application of cross immunity, or differential serum virus neutralization tests. Those attempting work in this field should be thoroughly familiar with the American Public Health Association's Diagnostic Procedures for Virus and Rickettsial diseases.[5] The intracellular structures sometimes known as inclusion bodies were once thought to be of great diagnostic significance, but with the exception of the negri body test for rabies this has not been borne out in practice. Guarnieri bodies are observable in biopsy specimens under optimal conditions. According to Weinstein[6] this test was reported "negative" on the original suspect in the 1947 outbreak of smallpox in New York City. It is possible to demonstrate intracytoplasmic or intranuclear inclusions in infections of the chorio-allantoic membrane of the developing egg. Demonstration of rickettsiae and some of the larger viruses stained by the Macchiavello, Giemsa, or other technics is possible but requires experience. The utilization of the electron microscope in the identification of viruses strictly concerns research procedures.

When the identity of the virus has been established, proof of its origin from the patient rather than the laboratory animal not infrequently must be established, particularly if the laboratory animal has at any time been shown to be a reservoir of the virus in question. The embryonated egg does not always escape suspicion. In establishing the role of the virus in the patients' illness specific serologic evidence, in conjunction with clinical, epidemiological and other evidence may require consideration by the physician.

Serological Tests With Specific Viral and Rickettsial Antigens As An Aid in the Identification of Viral and Rickettsial Diseases

It must be emphasized that the serological identification of viral diseases has a wider field of practical application than the approach through the attempted recovery and identification of the suspected virus. Many of these tests can be done in the well managed clinical or public health laboratory which already perform the complement-fixation test for syphilis. The test tube procedures with specific viral and rickettsial antigens recommended are limited principally to the complement-fixation and hemagglutinin-inhibition technics, since these procedures involve utilization of killed antigens. Since the antigens are expensive some laboratories prefer the utilization of their own antigens, but it must be admitted that the cultivation of viruses for antigen production is tedious, troublesome, and with many agents, a dangerous procedure. It must be admitted that both the physician and patient frequently lose interest in serological tests because the height of the illness may have been reached or passed before a significent rise in titer was detected. While

the necessity of testing two or more specimens perhaps cannot be avoided, nevertheless late information can be of great help to the physician in making a retrospective diagnosis.

Other specific serologic procedures with less limited application include rickettsia agglutination and serum virus neutralization. Rickettsia agglutination cannot be done with the soluble antigens utilized in the complement-fixation test. The serum virus neutralization test is expensive and time-consuming and since it requires the inoculation of embryonated eggs or mice, it is not recommended for the clinical laboratory routine. The serum virus neutralization test continues to be of great value in the virology research laboratory and it frequently happens that this is the most important serology procedure available.

The complement-fixation test is by far the most widely useful procedure available in the recognition of viral and rickettsial diseases. The Kolmer or similar procedure with the usual reagents with substitution of the specific viral or rickettsial antigen is suitable. Attention must be accorded technical details in titration of each reagent in a standard procedure. Known positive and negative serums must be included with each test. It is advisable to test paired serums from the patient simultaneously with a battery of antigens, or to make use of a screening procedure. Problems frequently arise concerning the sensitivity or specificity of tests. The greatest source of difficulty concerns the antigens, but maintaining suitable "positive" and "negative" control serums is at times a problem. Antigens oftentimes are of low potency or possess poor keeping qualities. Crude home made antigens especially tend to give poor results, reacting with syphilitic serum or non-specifically with "negative" serum. Cox[7] and his collaborators have been particularly active in elucidating the principals underlying the production of viral antigens and vaccines. A number of specific viral and rickettsial antigens prepared from infected mouse brains of embryonic tissues or fluids and embryonated egg yolk sacs respectively are commercially available and include: Antigen control (normal tissue), Colorado tick fever, equine encephalomyelitis (Eastern) (EEE), equine encephalomyelitis (Western) (WEE), influenza A, influenza B, lymphocytic choriomeningitis (LCM), lymphogranuloma venereum (LGV), mumps, normal mouse brain control, psittacosis-human pneumonitis, Q fever, rickettsialpox, Rocky Mountain spotted fever, St. Louis encephalitis, screen antigen for spotted fever group, typhus (epidemic) and typhus fever (murine). Most of the "positive" control serums are also available from commercial sources. Our experience with several of the commercial viral antigens has been limited. The results obtained must be reported cautiously.

The titration of antibodies for some viral diseases is possible by means of the hemagglutinin-inhibition reaction first applied by Hirst[8] in the study of influenza. The test depends on the fact that certain viruses agglutinate erythrocytes. Chicken red blood cells are generally utilized. Since serum antibody inhibits virus agglutination of suitable erythrocytes, antibody titration is possible by use of serum dilutions and a constant dose of virus and erythrocytes. This test is restricted mainly to influenza and mumps. The inhibition titers are read in serum dilutions similar to those used in bacterial agglutination tests. As with the complement-fixation test, it is frequently impossible to assign significance to titers obtained on a single test because of distribution of titers found in random samples. Nothing less than a four-fold titer increase late in or after the illness is considered significant. To the extent it is applicable, hemagglutinin-inhibition offers a check on the results to the complement-fixation test.

Non-specific Tests As An Aid in the Identification Of Viral and Rickettsial Diseases

Several tests, of some value in the recognition of viral or rickettsial disease, are performed with non-viral antigens. Perhaps the most widely used are the variations of the Weil-Felix test with proteus OX antigens in the recognition of rickettsial diseases; the heterophile test for infectious mononucleosis; and the cold agglutinin and the streptococcus MG agglutination tests for primary atypical pneumonia.

The Weil-Felix agglutination test utilizing proteus OX-19 antigen formerly was widely employed in the recognition of typhus fever, and variations of the test with additional antigens were advocated in the differentiation of typhus and spotted fever. These procedures now are known definitely to be inferior to the complement-fixation procedure utilizing specific typhus and spotted fever antigens. The complement-fixation tests are much more sensitive and specific for typhus and spotted fevers. It has not been possible to use the Weil-Felix test in the recognition of Q fever.

Since the properties of the virus of infectious mononucleosis have not been determined, the specific viral antigen is not available, and the heterophile test continues to be widely utilized. It must be emphasized that this test is essentially non-specific. It is less non-specific when the serum is absorbed with guinea pig kidney. The detection of a rising titer during or shortly after the illness has most significance.

Only non-specific serologic aids are available in recognition of primary atypical pneumonia. These are the streptococcus MG agglutination and the cold agglutinin tests. Curiously enough, there is much evidence that the more serious illnesses tend to result in the highest titers.

It is also a curious fact that while the streptococcus MG apparently is not the cause of primary atypical pneumonia, the patient's serum frequently agglutinates a suspension of this organism. The streptococcus MG is a non hemolytic streptococcus which appears to be distinct serologically from other species of streptococci. Low titers during the illness are viewed with extreme caution.

The cold agglutinin test is based on the fact that serum from the case of primary atypical pneumonia tends to agglutinate human group O erythrocytes in the cold room. At 37° C the agglutination disappears. There seems to be little agreement concerning the optimal concentration of cells, although perhaps .2% is preferable. The serum to be tested for cold agglutinins should be warmed at 37° C before the clot is separated, particularly if the specimen has been in the refrigerator. Titers of 32 or more may be considered abnormal. The demonstration of an increase in titer during the illness is most significant. While the evidence is conflicting there is some evidence that the cold agglutinin test is more sensitive than the streptococcus MG agglutination test for primary atypical pneumonia.

Table I

Approximate Number of Tests for Virus and Rickettsial diseases done by Texas State Department of Health Laboratories in 1948

VIRUS RECOVERY TESTS		SEROLOGICAL TESTS	
Total Tests	150	Serum Virus Neutralization Tests in Eggs or Animals	25
Positive Findings	9	Virus Hemagglutination Inhibition Tests	75
Smallpox	6	Virus Complement Fixation Tests	250
C Virus	2	Rickettsial Complement Fixation Tests	5000
Herpes	1	"Non-specific" Tests	3000
Rabies	0		

Tests for Viral and Rickettsial Diseases in the Texas State Health Department Laboratories

In the Texas State Health Department Laboratories most emphasis is put on serological tests in assisting physicians with the recognition of viral and particularly rickettsial diseases, as shown in Table I. Commercial antigens are utilized exclusively in doing rickettsial tests, mainly for typhus and Q fever. Both commercial and home made antigens are utilized in the performance of complement-fixation and hemagglutination inhibition tests for viral diseases. Our experience with preparation of satisfactory viral antigens has concerned largely frozen and dried benzene extracted mouse brain antigens somewhat similar to some of the commercial antigens. Relatively few in vitro serum virus neutralization tests are done. Several research projects have been attempted in recent years. A considerable num-

ber of tests for typhus fever on rats and Q fever tests on other rodents and domestic animals is not shown in the tabulation. It is of interest that the first naturally acquired, sizable outbreak of Q fever acquired in the United States was identified on the basis of complement-fixation tests performed in this laboratory.[9] In 1949, notable findings concerned illnesses which occurred late the previous year in a poultry and egg plant. High titers[10] which gradually decreased over a period of several months were obtained with Lygranum antigen and aided in the identification of an outbreak of psittacosis probably resulting from working with turkeys in the poultry and egg plant.

Status of Laboratory Tests for "Neurotropic" Viral Infections Including Poliomyelitis

While much progress has been attained in devising specific viral antigens for use in complement-fixation tests which aid in the recognition and differentiation of the neurotropic viral diseases, including St. Louis encephalitis, Western and Eastern equine encephalomyelitis, lymphocytic choriomeningitis, and the Coxsackie group of viral diseases uncovered by Dalldorf and co-workers,[11] it must be emphasized that there is no practical, satisfactory test for poliomyelitis. Recent typings suggest that the great majority of poliomyelitis virus strains fall in three or four immunogenic groups. Likewise the recent demonstration by Weller, Robbins and Enders[12] of a method of cultivation of poliomeyitis virus in vitro utilizing nonnervous human tissue suggests that satisfactory serological tests eventually will become available. The quest for poliomyelitis virus in the human stool or elsewhere remains essentially a research procedure which necessitates the inoculation of the monkey.

In the differentiation of non-paralytic poliomelitis and Coxsackie disease, the search for the latter virus in blood, stools and other sources is assuming increasing importance. The infant mouse is susceptible to the "C" virus of which at least eight immunogenic groups already are known. When the "C" antigens become more generally available laboratory aids to the physician in the diagnosis of this disease will assume more importance.

Summary: The recovery and identification of viruses involve procedures which are largely in the realm of research. Serological procedures, utilizing specific viral and rickettsial antigens, capable of yielding results of value to the physician in the identification of viral and rickettsial diseases, are suitable for use in many medical and public health laboratories. Non-specific serological procedure are less useful. The application of some of these tests in the laboratories of the Texas State Health

Department was discussed. The lack of a satisfactory test for poliomyelitis was noted.

REFERENCES

1. Barnard, Chester I., The Rockefeller Foundation: Review of work in 1949, New York.
2. Smadel, J. E., The Practioner and The Virus Diagnostic Laboratory, J. A. M. A. 136:1079-1081, 1948.
3. Bohls, S. W., and Irons, J. V., Chorio-allantoic Membrane Infection As a Diagnostic Test for Smallpox, Amer. J. P. Health, 32:300-306, 1942.
4. Cox, Herald R., Use of Yolk Sac of Developing Chick Embryo As Medium for Growing Rickettsiae of Rocky Mountain Spotted Fever and Typhus Groups, Pub. Health Rep., 53:2241-2247, 1938.
5. Francis, Thomas, Jr., with 18 collaborators, Diagnostic Procedures for Virus and Rickettsial Diseases, 1st Edition, American Public Association, 1948.
6. Weinstein, Israel, An Outbreak of Smallpox in New York City, Amer. J. P. Health, 37:1376-1384, 1947.
7. Cox, Herald R., Specific Complement-Fixing Diagnostic Antigens for Viral and Rickettsial Diseases. Amer. J. P. Health, 38:351-360, 1948.
8. Hirst, G. K., The Quantitative Determination of Influenza Virus and Antibodies by Means of Red Cell Agglutination, J. Exp. Med., 75:49-64, 1942.
9. Irons, J. V., Murphy, J. N., Jr., and Wolfe, D. M., Q Fever in the United States III. Serologic Observations in an Outbreak Among Stock Handlers and Slaughterhouse Workers, J. A. M. A. 133:819-820, 1947.
10. Irons, J. V. and Sullivan, T. Unpublished observations.
11. Dalldorf, G., Sickles, G. M., Plager, H., and Gifford, R. A., Virus Recovered from the Feces of "Poliomyelitis" Patients Pathogenic for Suckling Mice, J. Exp. Med., 89:567-581, 1949.
12. Weller, T. H., Robbins, F. C. and Enders, J. T., Cultivation of Poliomyelitis Virus in Cultures of Human Foreskin and Embryonic Tissues, Proc. Soc. Exp. Biol. and Med., 72:153-155, 1949.

A TECHNOLOGIST'S VIEWPOINT OF THE COMMUNITY AND WALKING BLOOD BANKS*

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The purpose of this paper is to acquaint you with the organization and type of work being carried on in the Danville Elks Blood Bank. At the time this paper is being written the bank has been in operation for a period of three years. However, this paper will cover the first two and a half years of operation. The personnel of the bank throughout this period has at all times endeavored to serve the hospitals, doctors, patients and blood donors with the finest type of professional service possible. Throughout this period of operation the bank has been a success from both a financial and professional standpoint. We have encountered problems, some of which would be found in any organization of this character, others are of a particular local

* Read before ASMT convention, June, 1950, Houston, Texas.

nature. We feel all these issues are of interest to our professional colleagues.

The Elks Blood Bank about which this paper is written is located in Danville, Illinois, and derives its name from its financial sponsor.. The designation of the name seems appropriate, but we have found it would have been wiser to have left the sponsor's name out and chosen another equally as appropriate. This is the first point of interest to call to your attention if your community would be fortunate to have such a sponsor as ours.

Danville, Illinois, is loctaed due south of Chicago 140 miles by rail. A study of your map will quickly draw your attention to the fact that there are no other towns in this area the size of Danville for some distance. The surrounding country is made up of small communities in a farming and coal mining section of the state. The town of Danville has a population of 45,000.

The bank now serves the two local hospitals with all the whole blood, plasma and red cells that they request for transfusion work. We have been carrying on this service since the bank started operation. Whenever the United States Veterans Hospital, which is located in the town, has called on the bank for blood we have been able to serve them. Whenever citizens of the town have received professional care in other medical centers and have been in need of whole blood, and these centers will accept our product, we have served them. Through this gesture the bank has been able to extend its services outside its regular territory. We are looking forward to a time when an exchange system can be put into effect where rare types of blood can be exchanged. There have been times when we have had to convert from six to eight units of Rh negative blood into plasma. This seemed to be a waste of Rh negative blood. With an exchange system possible, other communities could have benefited by its use.

In the past year the bank has been visited by the following persons: Dr. T. H. Seldon, now president of the National Association of Blood Banks; Dr. G. Albin Matson of the War Memorial Blood Bank at Minneapolis; and most recently by Dr. Malcolm MacEeachern of the American College of Surgeons. Each of these well known persons was pleased with the organization and the type of professional service they found being carried on in the bank.

How did the Danville Elks 332 Blood Bank come into existence? When the medical men of the community returned from service, they felt that without a blood bank the future of medicine in the community would be greatly impaired. The project of a blood bank for their community was presented to the County Medical Society. One doctor was appointed to

make a study of blood banks nad visit some of those in operation. Another was appointed to secure a financial sponsor. These men reported to the Society that their community could support a bank, and that a financial sponsor had been secured.

The Society appointed five medical men to comprise the Blood Bank Board, and one of the committee was appointed Medical Director. This Board is responsible for the operation of the bank. Up to this date the town has not been serviced by a full time pathologist. Since the first of the year, however, this situation has been changed. One hospital has a full time pathologist and the other hospital is served by a part time pathologist. We are all aware that our Clinical Pathologists are the persons who should be the supervisors of these technical projects, regardless of the Medical Boards that are so very necessary to a well functioning bank.

Why was the Elks Lodge chosen to sponsor the bank? The reason is that their membership is comprised of men from all races and religious creeds. The members were greatly impressed with the plans of the project and volunteered their unanimous support. This was the first blood bank in the United States to which the fraternal order of Elks had given complete financial support. For this reason, the bank has been visited by several Grand Exalted Rulers and other national officers.

The blood bank is a true community project. The local daily paper has given us their wholehearted support, and since it has a circulation of 50,000, this is no small advertising project. A staff photographer reports to the bank at the time every five hundredth donor is bled. The donor's picture along with an article of interest appears in the news the following day. The paper has also appealed for donors for various patients. Another wonderful help for the bank has been the local Radio Station WDAN. They have planned programs and broadcasted these in the interest of the bank, and at all times will appeal over the air for blood donors. The station has a listening area of over 100 miles. Since it is the only station for some distance, one can readily understand why the bank can rely on the wonderful response we receive through their services.

Where should a small community bank be located? This is one of the problems that will present itself in any community. To be located in a hospital has its pros and cons. It does seem that the ideal location is away from the institutions that it serves. I have been in charge of two community banks located in hospitals, and the same problems have arisen in both situations. When the Elks committee faced this problem, the only available space was two rooms in one of the hospitals. This location was accepted by the committee as a home for the bank. One can readily understand that being located in one hospital

and serving another will require frequent adjusting in public relations that otherwise would not occur. It seems that opinions differ regardless of how hard the personnel of the department tries to serve all institutions on the same basis.

The bank was assigned to the two above-mentioned rooms. The larger is 15 by 15 feet. Here we receive donors, take histories, keep records, bleed donors and store the blood. Our two donor tables, which were designed and constructed under my supervision, are arranged here also. They are separated from the rest of the room by curtains, which assure the donor privacy while being bled. The adjoining room is 8 feet wide by 15 feet long. In this space all the technical work is carried on.

All new equipment was purchased for the department, and as it was of the very best, it will be some time before any will have to be replaced.

From my experience in community banks, I would suggest that endowment funds be greatly encouraged. These gifts will give the bank a sound financial future.

The balance of this paper will cover the first two and a half years of operation. The reason for choosing this exact period is that we bled 4000 donors. The first 25 were all volunteers from the Elks Lodge, after which we operated on replacement donors only. We are often asked how can this be done? This is the way it has been handled. For the first three units of whole blood dispensed to the same patient, the bank requires two replacements. After the third unit, the requirements are dropped back to one donor for each unit. In the case of plasma, one donor for each unit of 300 cc is required. When washed red cells are dispensed, this requires one regular replacement donor per 250 cc.

Some seasons of the year the bank will fall behind in replacements. With the aid of the radio and press, there has never been a time when a patient needing blood did not receive it at the time it had been ordered. If we were to press collections on all accounts with either replacements or money payments, the bank would never fall behind in blood or finances. Since the bank has been doing an average of nearly $6000 per year of charity work, one can understand how this situation would arise. Of course this amount of charity work is made possible only through the generosity of our financial sponsor.

Another project of interest will be our "Walking Blood Banks." These were started at the time the bank began operation. Various lodges, PTA's, industrial groups and civic clubs were contacted. At each of these meetings the technologist of the bank spoke to the group. Blood typing and Rh determinations were made on all who volunteered to place their names on a donor list for the organization. Each volunteer was pre-

sented with a donor card with his name, address, blood type and Rh determination. These cards stimulated interest, and others volunteered. Through this gesture the "Walking Blood Banks" were started. Each group selected a chairman to head its donor list whose duty is to keep all records, secure replacement donors, and make appointments for donors. It is well to select a community spirited chairman, entirely acceptable to the area, to head each "Walking Blood Bank."

The present age limit for our donors is as follows: males 18 to 60, females 21 to 60. Some may not agree with this range of ages. However, in screening donors there is a personal factor that does enter in.

When a unit of whole blood, plasma or red cells has been dispensed to a patient, a charge card with an envelope containing two donor instruction cards is left at the bedside of the patient to be presented to the nearest relative, who in turn contacts the donors. The instruction cards contain a prescribed diet to be followed for at least ten hours prior to their visit to the bank. The importance of consuming a large amount of water is stressed during this ten-hour period, and as a result we have very few dehydrated donors. We rarely have donors who become ill. Any number of donors report they do not drink water and have had unpleasant experiences as blood donors, while the same donors after following our instructions rarely notice any change in themselves.

The blood donor must be in reasonably good health. The blood pressure must fall within the range diastolic not below 100 and systolic not over 200. Temperature reading must not read over 99 degrees. A hemoglobin reading is made on all donors before each donation. The method used is Lumetron Photoelectric. Persons who do not give a reading of 14.4 grams, or around 84.5 per cent, are rejected. At the time blood is taken for the hemoglobin, a drop is dropped into a copper sulphate solution which has a specific gravity of 1.052. The drop of blood is carefully observed as it falls through the solution. If it is slow or hesitates before falling, this observation is correlated with the hemoglobin reading and is helpful in screening donors. No one person is allowed to donate more than every three months. Persons are rejected when they give a history of having had the following diseases: tuberculosis, malaria, jaundice, diabetes, heart trouble, rheumatic fever, syphilis, and those having undergone brain surgery. There are other illnesses which prohibit one from donating, but most of these are on a temporary basis and will not be mentioned here.

The Baxter System is used throughout the bank. The bleeding vacs are the 600 cc size which contain 120 cc of ACD* solution.

* ACD solution—Each 100 cc contains: 2.45 gms. Dextrose, U. S. P., 1.37 gms. Sodium Citrate, U. S. P., 0.50 gms. Citric Acid, U. S. P.

Disposal donor sets are used. We stock these sets with the 17 and 18 gauge needles, the use of which is determined by the size of the donor's vein. A drop of 2% Procaine is injected at the site selected to inject the donor needle. At this time, the donor is handed a piece of gauze saturated with aromatic spirits of ammonia. He is instructed to smell this while the blood is being drawn to lessen the possibility of fainting. The operator is instructed to engage the donor in a conversation that does not have any phase on blood drawing. The bleeding room is kept as cool as possible. All overhead lights are off. As soon as the phlebotomy is over, the donor chooses a drink from the menu. Snacks are not served simply because we do not have the room to prepare them. The donor must rest 20 to 30 minutes before leaving the bank.

When the donor has consumed sufficient water, we have found it takes from three to five minutes to draw a unit of blood with a 17 gauge needle, and five to eight minutes with an 18 gauge.

To avoid any mixup in units of blood, I have developed the following method for storing red cells and serums. Two Wasserman tubes are strapped on the side of the bleeding vac at the time it is placed in the refrigerator. These are corked. When the phlebotomy is over and before the needle is removed from the donor's vein, the valve needle is removed from the vac and 3 cc of blood is run into one tube, the other one is filled to be used for serum. To the 3 cc of whole blood several drops of ACD solution is added before the blood clots. This tube is never removed from the vac while it is stored in the bank. Samples are taken from this tube for the making of cell suspensions used in blood typing and compatibility tests. The blood remaining in the donor tubing is used for making the Rh determinations and for the serology tests. 5 cc of the serum is strapped on the vac beside the tube containing the red cells. This will be used in cross-matching technics.

All donors are typed with A,B and O typing sera, using a test tube technic. An Rh determination is made on each donor using the slide technic. All true negative results are checked with the test tube method. We are using anti-c, anti-D, anti-CD and anti-E seras for these tests.

The standard Kahn test is run on each blood. Most recently the VDRL antigen technic has been installed as routine. All bloods giving a positive serology are discarded at once.

Some interesting facts were observed from a study of the records made on the 4000 donors. The donors were divided into the following age groups: Males: 18 to 30, 31 to 40, 41 to 50, 51 to 60; females 21 to 30, 31 to 40, 41 to 50, 51 to 60. The

total male donors was 3433, female donors 567. The two age groups where the greater number of donors fell were males 18 to 30, 31 to 40.

Male	Number	Female	Number
18-30	1566	21-30	205
31-40	1021	31-40	140
41-50	561	41-50	129
51-60	287	51-60	93

Wherever transfusions are being administered, the one concern of all is how to keep reactions at a minimum. I have emphasized the diet of the blood donor; this has given interesting results. During the period we bled our 4000 donors, we dispensed 2953 whole bloods, 285 plasma, and 16 washed red cells for transfusion work. The number of reactions reported to the bank was 21. Upon investigation of these it was found that 17 were diagnosed by the physician as of some allergenic source. The remaining three were not determined. One reaction from plasma, and when this was checked we found that regardless of what was given theis patient intravenously a reaction followed.

The bank has been called upon to do most of the compatibility work for the patients in the hospital where the bank is located. Routinely a major and a minor cross-match is done. Technic used is a test tube method. Blocking antibodies and cold agglutinins are checked. A sensitive cross-match is done on all patients that have received transfusions within a period of several weeks. The same test is made on all obstretical cases.

The bank stores all whole blood for 21 days. After the exploration date the plasma is aspirated into pooling vacs (2000 cc) containing 200 cc of 50% dextrose solution to prevent the formation of fibren. When the pooling vac is filled, 30 cc of plasma is secured for aerobic and anaerobic cultures. Fluid Thioglycollate media is used for all culture work. The cultures are incubated at 37.5° C. for 14 days before negatives are reported. After the sample is removed for cultures, 20 cc of a 1% solution of "Merthiolate" is added for a preservative. The pools are stored at room temperature in a dark cupboard. When the plasma is to be dispensed, it is aspirated into 300 cc vacs. Again cultures are made and observed for seven days. If the cultures are negative, the units are ready to be dispensed.

The bank has been called upon several times to answer emergencies in other communities. First was the Centralia mine disaster, and again we were asked to stand by at the time of the Effingham Hospital fire. It is our policy to have on hand currently 50 units of plasma in readiness for such emergencies.

One all important suggestion to make is first keep the tech-

nical work of the bank on a high professional level. Keep the bank operating on a high community level, never allowing one or several of any group to gain monopoly of the project.

Spend time in choosing a proper name for the bank. An incorrect name can be a stumbling block.

Arrange your financial setup so there will be no need of changes for some time. Above all else, have the same system working throughout the community.

SUMMARY

A community of 45,000 population can support and operate a bank successfully from both a financial and professional standpoint.

Well organized "Walking Blood Banks" are most helpful in securing replacement donors.

A bank can operate on replacement donors, if the doctors, hospitals and relatives of the patient will cooperate.

Endowment funds will strengthen the bank for the future.

Well chosen diets for donors will help reduce reactions in the patients receiving their blood.

When donors are not dehydrated, it lessen reactions in them.

In national disaster or in any great emergency where lives are in danger, a blood bank is the first line of defense.

Medical Technologists have a vital role to play in this all important field of blood banks.

THE VALUE OF AN INSULIN TOLERANCE TEST IN THE DIAGNOSIS OF FUNCTIONAL HYPOGLYCEMIA*

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The purpose of this paper is to discuss the advantages of an insulin tolerance test in the diagnosis of functional hypoglycemia. We feel that in order to appreciate better the value of any test one must first thoroughly understand the mechanism of the principle upon which the test is based. So with this end in view, before presenting the test itself, we will briefly review carbohydrate digestion and absorption and discuss some of the most outstanding conditions which may give rise to a spontaneous hypoglycemia.

Ingested disaccharides and polysaccharides are converted into monosaccharides by the action of enzymes present in the salivary, pancreatic, and intestinal secretions, glucose being the most important end product. These monosaccharides are absorbed to a very small extent by the stomach and colon and

* Read before ASMT convention, June 1950, Houston, Texas.

to a greater extent by the small intestines and are taken to the liver by means of the portal circulation. It is believed that all monosaccharides are transformed into dextrose before being converted into glycogen. The dextrose brought to, or formed in the liver may undergo the following changes: 1. It may be stored as glycogen; 2. A small portion may be utilized directly in the metabolic activity of the liver cells; and 3. That which is not stored as glycogen or utilized directly by the liver, passes into the general circulation and is carried to the tissue, where it is either transformed into glycogen and stored as such in the muscles or utilized for energy or transformed into fat. The function of the glycogen stored in the liver is to maintain the normal blood sugar level; the function of that stored in the muscles is to furnish glucose for combustion in the tissues. Muscular contraction is associated with the conversion of muscle glycogen into lactic acid, about one-fifth of which is completely combusted into CO_2 and H_2O; the greater part of the remaining four-fifths gets into the blood stream and from there goes to the liver where it is again converted into glycogen; consequently, there is a regular cycle kept up which may be illustrated as follows:

Liver glycogen ⟶ Blood sugar ⟶ Muscle glycogen ⟶ Blood lactic acid ⟶ Liver glycogen.

The glucose content of the blood in a normal individual remains remarkably constant. A number of factors enter into control of blood glucose at a normal level of from 80 to 120 mg. % (Folin and Wu).

These factors may be listed as:

1. Formation of glycogen in the liver.
2. Formation of glycogen in the muscle.
3. The oxidation of carbohydrates.
4. Conversion of carbohydrates to fats.
5. Excretion of glucose.
6. Formation of glucose from amino acids.
7. Action of insulin.
8. Action of adrenalin (epinephrine).

The pancreatic hormone, insulin, tends to increase the oxidation of carbohydrates and to decrease the breakdown of glycogen. On the other hand, adrenalin increases the breakdown of glycogen and increases the production of lactic acid in the muscles

Spontaneous hypoglycemia may be defined as a condition in which the sugar content of the blood becomes markedly decreased. When this happens, a variety of symptoms may develop: hunger, sweating, weakness, trembling, anxiety, mental confusion or, if very severe, the patient may lapse into a coma or may experience generalized convulsions. If the presence

of a hypoglycemia is not proven, any one of a variety of erroneous diagnosis may be made; among these are epilepsy, intracranial tumor, psychosis, psychoneurosis and gastro intestinal disorders. The Medical Technologist can render valuable service by aiding the physician to arrive at a correct diagnosis.

According to Dr. Conn[4] spontaneous hypoglycemia or rather the causes of spontaneous hypoglycemia may be classified as follows:

I. Organic (with recognizable anatomic lesions)
   a. Pancreatic tumors.
   b. Hepatic diseases.
   c. Pituitary hypofunction.
   d. Adrenal hypofunction.
   e. Central nervous system lesions.

II. Functional (without recognizable anatomic lesions)
   a. Post alimentary and exertional.
   b. Hypoglycemia of pregnancy and lactation.
   c. Renal glycosuria.
   d. Autonamic imbalance.

III. Miscellaneous
   a. Factitious.
   b. Postoperative hypoglycemia.
   c. Severe inanition.
   d. Unknown.

The organic type of hypoglycemia, in general, does not present a very difficult problem as far as diagnosis is concerned because of the fact that the fasting blood sugar is consistently low, and there are usually anatomic lesions that may easily be discovered by X-ray investigation or laboratory tests.

Functional hypoglycemia according to Conn[3] is by far the most common. It accounts for at least 70 per cent of all cases of spontaneous hypoglycemia. There is quite a bit of controversy as to what this syndrome should be called, which matters little so long as it is recognized for what it is and treated accordingly. The fact that the fasting blood sugar may be normal and there are usually no anatomic lesions renders the diagnosis of this type of hypoglycemia a difficult problem.

It is an universally accepted fact that a diagnosis of any type of hypoglycemia is definitely established if an abnormally low blood sugar is present during the attack, and the symptoms are immediately relieved upon the administration of glucose. Unfortunately, however, the severe attacks in functional hypoglycemia are usually of very short duration because of prompt and vigorous counterreaction, and it is difficult to obtain blood for sugar determination promptly enough before these counterreactions have altered the picture.

The prolonged glucose tolerance as a diagnostic aid is not

always reliable because of the variety of curves that may be obtained even in the same person. Cantarrow[1] maintains that the glucose tolerance must often be repeated before apparently abnormal results can be interpreted satisfactorily and that false normals or borderline normal curves are sometimes obtained as a result of gastric disturbances. For example, in reviewing a series of 50 prolonged glucose tolerance tests, which we performed in our laboratory, using a standard dose of 100 grams of dextrose and making blood sugar determinations every hour for six hours, we found that only 10% showed positive hypoglycemic curves, (blood sugar at the end of six hours below 70 mg. %), while 30% showed borderline normal curves. Since we have no data on these patients other than the above test, we cannot use them as a criterion in evaluating a prolonged glucose tolerance as a diagnostic procedure for hypoglycemia; nevertheless, we may safely presume that the majority, if not all, of these patients showed some clinical manifestations of a hypoglycemic state since a six hour and not a shorter procedure for glucose tolerance was requested.

The use of an insulin tolerance test in the diagnosis of a spontaneous hypoglycemia became feasible with the introduction of insulin as a thereapeutic agent in the treatment of psychosis. The principle of the test is based on the reaction of an individual to an insulin induced hypoglycemia. Normally the body is able to set up a series of counterreactions which will destroy the effect of insulin and restore the blood sugar concentration to a normal level, but the individual with functional disorders will be unable to elicit these counterreactions with the result that the hypoglycemic stage will be prolonged.

We have found the following procedure for insulin tolerance test of great value in aiding the physician in the diagnosis of functional hypoglycemia. The patient is allowed nothing by mouth after 9 P.M. A fasting blood sugar is obtained, and then a single dose of 10 units of regular insulin is given subcutaneously, and samples of blood for sugar determinations are drawn at thirty, sixty, ninety, and one hundred and twenty minute intervals. The findings in 33 normal individuals are given in Table I.

The subcutaneous injection of a standard dose of 10 units of regular insulin rather than the more widely used intravenous dose of 0.1 unit per kilogram of body weight needs some explanation. The principal of the test is primarily based on the ability of the body to restore its diminished sugar concentration to the normal level, which will be possible only if there is an adequate supply of available glycogen in the liver and muscles. The excess weight of an individual is due to adipose tissue, which is composed of fat and does not bear any imme-

Table I

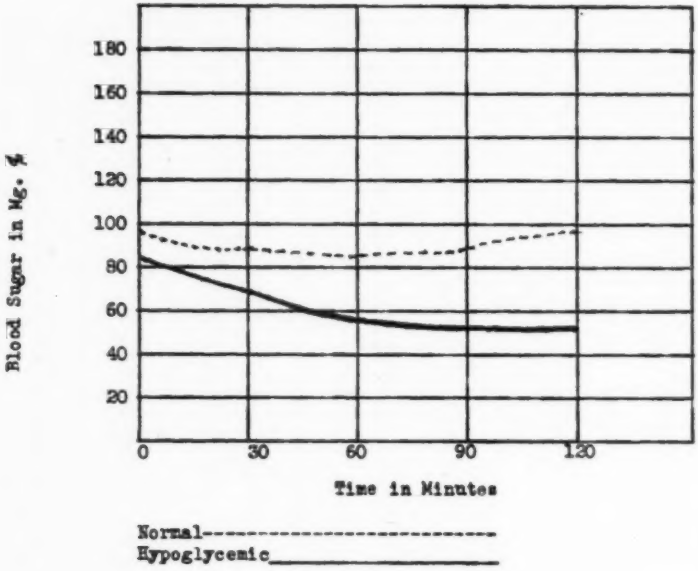
Findings of Insulin Tolerance in 33 Normal Individuals

CASE No.	BLOOD SUGAR MG. PERCENT				
		Minutes After Injection of Insulin			
	Fasting	30	60	90	120
1	97	92	93	93	92
2	100	88	85	87	95
3	105	100	88	90	98
4	82	73	72	78	90
5	117	86	73	90	96
6	95	82	75	78	89
7	110	95	83	84	105
8	91	82	84	86	96
9	100	92	85	84	97
10	115	114	111	100	112
11	84	67	70	83	85
12	111	107	78	85	102
13	110	88	85	95	98
14	115	107	100	97	110
15	86	73	82	90	90
16	95	82	75	78	88
17	105	100	96	101	107
18	94	93	82	91	97
19	90	87	85	85	84
20	89	90	86	84	90
21	105	100	100	95	101
22	135	111	108	96	120
23	88	85	86	86	91
24	85	75	73	86	93
25	95	90	76	87	99
26	90	91	84	83	95
27	79	76	74	85	89
28	96	94	90	90	95
29	88	82	83	90	89
30	93	91	78	88	96
31	95	90	88	87	94
32	99	102	85	92	100
33	106	95	93	96	98

diate relationship to the quantity of available glycogen; therefore, it follows that the body weight should not necessarily influence the mount of insulin given in this test. When insulin is given intravenously, the onset of action is immediate, but the maximal effect is short-lived since the hormone is rapidly destroyed and excreted. We found that when given subcutaneously the maximal effect of 10 units of regular insulin was reached in 30 to 90 minutes which eliminates the hazards of a sudden and violent reaction in patients suffering from severe hypoglycemia. Our normal and abnormal insulin tolerance findings are illustrated by graph in Table II.

We found that taking the average of the normal group the blood sugar dropped about 13 mg. or 14 per cent in sixty minutes after the subcutaneous injection of 10 units of insulin but was back to preinjection level in from ninety to one hundred and twenty minutes. This group developed no reaction whatever to the insulin.

Table II
Graph Showing Normal and Abnormal Curves in Insulin Tolerance



In the group of 33 patients in whom clinical manifestations of hypoglycemic attacks had been observed the average fall in the blood sugar after the subcutaneous injection of 10 units of regular insulin was 16 per cent in thirty minutes, 31 per cent in sixty minutes, and 34 per cent in ninety and one hundred and twenty minutes. The complete findings for this group are given in Table III. Emphasis should be placed on the fact that the duration rather than the degree of hypoglycemia is the outstanding diagnostic characteristic of this test.

According to Dr. Helfer[8] all of these patients showed different improvement after they had been placed on a high protein diet, which is further evidence that their symptoms were the result of a hypoglycemic state. Conn[2] recommended the use of a high protein diet as a therapeutic measure in functional hypoglycemia because the time required for the absorption of protein and its conversion to dextrose affords a steady supply of glucose over a long period of time. Although protein yields 50 per cent of its weight as dextrose, it does not give rise to a

sudden elevation of blood sugar and a subsequent fall as does a diet high in carbohydrates.

The procedure we used for the insulin tolerance proved time saving, safe, and inexpensive. The patient need not be hospitalized but may report to the laboratory for the test, which can easily be completed in about two hours. A very small percentage of the hypoglycemic group developed a reaction to the insulin. This reaction varied from complaints of hunger, to dizziness, extreme weakness, slight mental confusion, and, in one or two cases, to profuse sweating and trembling. The test was completed in all cases, and the symptoms were immediately relieved by a glass of orange juice with some sugar added.

We are convinced that there is hardly any other test that gives such clear cut diagnostic results as does the insulin tolerance in the diagnosis of functional hypoglycemia. Therefore, we have taken this opportunity to present our experience with this test with the hope that others may investigate its diagnostic

Table III

Findings of Insulin Tolerance in 33 Patients in Whom Symptoms of Hypoglycemia Had Been Observed

CASE No.	BLOOD SUGAR MG. PERCENT				
		Minutes After Injection of Insulin			
	Fasting	30	60	90	120
1	97	95	53	44	46
2	95	67	51	51	44
3	88	78	53	53	56
4	105	53	59	62	65
5	127	110	80	52	59
6	66	62	54	52	52
7	75	73	62	62	59
8	82	82	73	59	56
9	85	78	67	65	65
10	88	67	51	59	62
11	67	50	42	41	43
12	73	60	56	60	60
13	80	54	60	60	62
14	112	95	60	63	60
15	77	70	67	62	67
16	74	51	46	44	49
17	88	75	49	52	56
18	73	64	50	50	51
19	70	67	53	46	51
20	53	46	41	44	44
21	78	73	70	59	51
22	82	73	62	59	56
23	97	92	52	56	65
24	89	53	50	53	59
25	90	87	75	59	58
26	82	79	73	63	57
27	70	57	57	50	63
28	85	71	72	70	60
29	85	76	54	53	57
30	79	53	52	48	46
31	83	67	65	60	64
32	75	50	40	41	38
33	90	84	50	51	53

potentialities and improve on it, so that it may become 100 per cent accurate in the diagnosis of this syndrome.

Summary

An insulin tolerance test, using a subcutaneous injection of a standard dose of 10 units of regular insulin, is reported, with tables showing complete blood sugar findings in 33 normal individuals and in 33 patients exhibiting clinical manifestations of functional hypoglycemia. Carbohydrate metabolism and the classifications and outstanding characterics of spontaneous hypoglycemia, with special reference to the functional type are reviewed.

REFERENCES

1. Cantarow, Abraham, and Max Trumper, *Clinical Biochemistry*. Philadelphia. W. B. Saunders Company, 1947. pp 1-58.
2. Conn, Jerome W., "The advantages of a High Protein Diet in the Treatment of Spontaneous Hypoglycemia," *Journal of Clinical Investigation*. 15: 673-78. November 1936.
3. Conn, Jerome W., "The Spontaneous Hypoglycemias," *The Journal of the American Medical Association*, 115: 1669-75, November 1940.
4. Conn, Jerome W., "The Diagnosis and Management of Spontaneous Hypoglycemia," *The Journal of the American Medical Association*, 134: 130-37 May 1947.
5. Duncan, Garfield G., *Diseases of Metabolism*. Philadelphia: W. B. Saunders Company, 1949. pp 657-77.
6. Greenblott, Robert B., and Herbert S. Kupperman, "The Diagnostic Value of the Glucose Tolerance and the Insulin Tolerance Test in Endocrine Disorders," *Southern Medical Journal* 40: 737-44 September 1947
7. Helfer Lewis M., "Hypoglycemic States," *Texas State Journal of Medicine* 39: 15-19. May 1943.
8. Helfer, Lewis M., Personal Communication.
9. Hyman, Harold Thomas, *An Integrated Practice of Medicine*. Philadelphia: W. B. Saunders Company, 1948. II, 1242-1246.
10. Maersch, Frederick P., and James W. Kernohan, "Hypoglycemia: Neurologic and Neuropathologic Studies," *Archives of Neurology and Psychiatry*, 39: 242-57, February 1938.
11. Nielson, Waldo E., *Textbook of Pediatrics*. Philadelphia: W. B. Saunders Company, 1950 pp. 1435-39.
12. Peters, John P., and Donald V. Van Slyke, *Quantitative Clinical Chemistry Interpretations*. The Williams and Wilkins Company. Baltimore: 1946.
13. Rector, John Mott and Robert E. Jennings, "Functional Hypoglycemia of Childhood." *American Journal of Diseases of Children*. 53:1012-21 April 1937.
14. Wilder, Russell M., and others, "Carcinoma of the Islands of Pancreas," *The Journal of the American Medical Association*. 84: 348-54, July 1927.

ORGANIZATION AND CURRICULUM SCHOOLS FOR MEDICAL TECHNOLOGISTS

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My subject is the school of Medical Technology; and I am assuming that it applies to those schools in our hospitals. Having been in close contact with the development of an "approved school," it may be that I can analyze it in a way that we can hope will be instructive to those who may be associated with a school and not too uninteresting to those who hope they never will be.

It is a school of Medical Technology and is allied to the Medical Profession; so let us first study in what way and manner this alliance exists. The American Society of Clinical Pathologists represent the group most vitally interested. Therefore this group has, for the Medical Profession, assumed the responsibility of establishing the standards of Medical Technology and registering those qualified. The American Society of Clinical Pathologists assumes no responsibility other than the qualifying of technologists for registration. The Council on Medical Education and Hospitals of the American Medical Association maintains the authority to qualify schools. The qualifying of the school of Medical Technology is done by the same person and at the same time as the hospital itself is visited and rated by the Council on Education for the A.M.A.

The Registry of the ASCP has specified to this Council on Education what they consider the essentials of an acceptable School for Clinical Laboratory Technicians. A booklet containing these essentials can be obtained from the Board of Registry. It is to be borne in mind that the Registry does not personally visit these schools listed as approved, but leaves it to the afore-mentioned representative of the American Medical Association. The visit of the representative of the A.M.A. is made at varying intervals. In the meantime a Pathologist may and frequently does outline his facilities and intentions thereby obtaining approval for his school pending inspection.

Before opening a school to train technologists, careful study is made of these suggested essentials. I have repeated the word "suggested" because both the American Society of Clinical Pathologists and the A.M.A. in listing the essentials of a school do not at any time say that by omitting any of the essentials a school will be closed. It is entirely a matter placed in the hands of the Pathologist concerned as to the type and manner of school he wishes to present as his contribution to educational advancement.

If one is in honesty to represent his laboratory as an adequate school, let us consider in detail what the student has a right to expect from such:

First, there must exist a department in every field covered in the education of a Medical Technologist, the minimum of which should be biochemistry, hematology, urinalysis, bacteriology, parasitology, histology, serology, blood bank, and record keeping. In each of these departments, there should be a registered technologist or otherwise qualified instructor.

Secondly, there should be a well-organized rotation of study in each department with examinations and practical experience.

Third, the hospital should have a variety of tests broad enough for the student to have information and instruction on any test in established use. By this I mean that one cannot say that a laboratory is a good place to train because it does 150,000 tests a year; for the judging cannot be by the number, but by the variety and the technique used in their execution.

And lastly, in addition to the academic value obtainable in the laboratory, it is the responsibility of the school of Medical Technology to be prepared to teach by example and discipline the professional dignity and efficiency essential to accurate work.

By summarizing as we have the expectations of the student, we can see what is required of a school worthy of the name "Approved." The solemnity of the undertaking is so great that one's mind turns to the words "not to be entered into unadvisedly or lightly."

Any hospital handling enough cases to consider the staff necessary for a school would probably have the departments listed. The additional need

in organizing the school is to have one person whose duty it is to have charge of this rotation of students and the teachers in each department; in other words, a chief technologist uniting the whole laboratory under one head; a person who, with the Pathologist, selects and checks the credentials of each applicant; who is responsible for the general management of the laboratory as a whole, which to be specific, would include the judging of the technical ability of the employed technologist and students, their attitude toward their work and professional associates, even to the minutest detail of their personal appearance. This person must have the authority and ability for this responsibility. No matter how conscientious and willing a pathologist may be, I know of no Pathologist in a position, or who wants to be in a position, to supervise students his entire day every day.

As I stated in the beginning, it is requested by the Council on Education that adequate records be kept of the laboratory work as a whole, the tests performed by each individual student, and the practice and lecture hours of each student. Every hospital has its own peculiar system of keeping records in its hospital and laboratory. I will point out certain essentials that I feel should be incorporated into whatever system is being used:

1. That the laboratory keeps in its files the original order received for work to be performed, and that on this same order shall be written the completed laboratory work with the signature of the person performing the test. This means that at any time the Pathologist may refer to any work in question and his investigation will show him the order that was received, the manner in which it was executed, and by whom.

2. Most all laboratories keep a record of the tests done each day in order to make correctly the charges to the patient's account. If this is not established, it should be so organized before a school of instruction is contemplated. This is accomplished by routine tabulation of tests and simple forms of bookkeeping.

3. The request of the Council on Education that a list of individual tests performed by each student be itemized was a stumbling block in our thoughts for some weeks. This has been solved by having each student daily record her activities on a work-sheet. This work-sheet is turned in each month. The chief technologist transfers this record to a form of monthly figures that enables a total for the student's year to be computed that will give the exact number of tests and also the variety performed. I might add that this also enables one at a glance to detect any tests that have not been performed by any given student as the period of her study draws to a close.

4. The record of attendance, practice, and lecture hours should be kept on each student as accurately and methodically as it would be kept in any institution of higher learning. This point on lecture hours brings out better than all else the need of schools of Medical Technology to be associated with a medical school which will permit the attendance of students in those classes pertaining to Medical Technology. For the working hospital to assume the responsibility of giving the necessary lecture hours and at the same time conduct efficiently a busy hospital laboratory is almost impossible. The students assume full responsibility for the subject and the examinations as given at mid-term and final. These grades obtained are recorded on their transcript together with the grade obtained by examination in each department of the rotating service. The number of hours spent in these formal lectures is recorded on a transcript and also those spent as practice hours in the laboratory.

No student should be accepted by a good school who has not previously had her transcript approved by the Registry. It is not safe for an institution of higher learning to accept a school as properly training Medical Technologists without a personal inspection and a close contact at regular intervals.

As time goes on, the student is constantly seeking a good school of Medical Technology that is affiliated with a college or university that in a four year period will give her three academic years and one hospital year with a degree and qualification for registry. The need today is not for more schools but for better ones.

A WORD FOR THE LABORATORIES*†

The reprint of an editorial in "Public Health Laboratory," a publication of the Conference of State and Territorial Public Health Laboratory Directors, in this issue of *Philadelphia Medicine,* should be given wider circulation because it is a very fair analysis of the article "MENACE" which appeared in the *Woman's Home Companion.*

It is to be hoped that superintendents of hospitals and others interested in the functioning of the nospital laboratories will read it seriously and give the laboratory directors a free hand in selection of their personnel.

No one, by the greatest stretch of the imagination, can say the directors have a free hand when the salaries are inadequate for the professional technical personnel needed in a modrn hospital laboratory.

In the writer's opinion, that is probably the crux of the situation.

The pathologist and clinical pathologist must have a sufficient number of well-educated and trained technical staff whose integrity is equal to their own. Those responsible for bacteriology, chemistry, hematology and serology must have assistants being trained for more responsibilities. It is hardly necessary to repeat that the salaries of such personnel should be decided by the director of the laboratory whose responsibility is to the physician, patient and hospital in that sequence.

Bacteriological technic is not acquired in one, two or three semesters; the same is true of chemistry and the other laboratory specialties. It takes years to develop proper procedures and technics.

The reprint of the editorial in *Public Health Laboratory,* appearing in this issue of *Philadelphia Medicine,* should be read and re-read. I feel sure that it will result in better selection and pay of laboratory personnel and encourage many of our younger people to devote their talents to hospital laboratories.

PUBLIC RELATIONS‡

A recent article by Albert Deutsch appearing in the *Woman's Home Companion* is titled "Menace in the Medical Lab." Most of the members of the Conference have probably seen it or had it called to their attention. In it you will find a somewhat jumbled account of mixed activities of public health and hospital laboratories interspersed with alarmist quotations from authorities in the laboratory field and citing selected examples of the shortcomings of laboratories in both public health and hospital backgrounds.

A Negative Approach

While we are most ready to admit that there is much room for improvement in the accuracy of laboratory work throughout the country, we deplore this negative approach to the problem. It is a fallacy to expect, as is inferred in this article, that the public health laboratory actually diagnoses diseases independently of the physicians through whom they carry out their proper function of providing factual evidence of bacteriological

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† Reprinted with permission from "Philadelphia Medicine," Sept. 9, 1950, Editorial at the request of ASMT Public Relations Committee.

‡ Reprinted from "The Public Health Laboratory," Bulletin of the Conference of State and Provincial Public Health Laboratory Directors, New York, May 1950.

*At the request of the Publication Committee, this comment was prepared by Claude P. Brown, M.D., Philadelphia, Penna.

and biochemical findings. The very first example cites the case of a patient from whom a blood specimen, probably a smear, was submitted and reported as containing malarial parasites; several months later the patient died from cancer, presumably because the laboratory failed to diagnose cancer in the blood smear which was examined for malaria. If cancer can be diagnosed from blood smears and if a patient cannot have malaria and cancer concurrently, it is news to us.

Entirely Different

In the CDC *Bulletin* for January 1950, Dr. M. M. Brooke made the statement: "In the current effort to eradicate malaria from the United States, epidemiologists and engineers are dependent upon laboratory examinations of blood films for definite proof of the existence of malaria in an area. Among 28,872 blood films examined by the parasitology laboratories of CDC between July 1948 and June 1949, only four were found positive for malarial parasites." In the article mentioned above we find the statement that "of twenty-eight thousand blood smears sent to CDC to *confirm suspected cases* of malaria only four were found to contain malarial parasites!" (The italics are ours.) Need we point out that confirming the presence of endemic malaria in a selected area and confirming suspected cases mean entirely different things in laboratory parlance.

But, perhaps this is all a needless quibbling as to details; our objection is to the emphasis on laboratory shortcomings and the entire lack of any mention of the many positive accomplishments of our public health laboratories and the services that they have been rendering throughout the years. Certainly such things as the declining attack rates of many communicable diseases, the control of epidemics and the increasing life-span of our population could not have been accomplished without the services of our public health laboratories. Hospital laboratories, also, have made notable advances and contributions during recent years; new biochemical tests, the provision of blood banks and Rh typing services could be mentioned as examples.

Agree on Lab Personnel

When we consider the statements of the Deutsch article in regard to laboratory personnel, we are in general agreement with all that he has to say. Many of our laboratory personnel *are* unscientific in their approach to problems, most of them *are* underpaid and overworked, and many of them are grossly underpaid for the responsibilities they bear. Such a result is the natural sequel to our present method of recruiting personnel. In hospitals we believe it it partly the result of the hospital's need to make money from its laboratory services, in many of them at least, by paying low salaries to unqualified "technicians." Until such a time as laboratory service is regarded as a profession and is raised to the level of other professions, such conditions are likely to persist. CDC among the states and several of the states among the private laboratories within the state are doing their best to raise the level of laboratory services through control of local laboratories, in-service training of local laboratory personnel, check specimens, etc., but it is a slow process, and at best can only be successful when we succeed in getting scientifically trained personnel to perform all scientific laboratory work.

Not a Menace

All in all, we cannot regard the present laboratory services as a MENACE to the public. We would like to see a more positive approach in presenting the facts to the public. Certainly, if any group of people need a good public relations counselor, it is the public health laboratory workers.

MAYBE IT WAS YOUR STATE SOCIETY THAT*

HAD A BOOTH AT THE STATE FAIR, and the Registered Medical Technologists typed 13,206 individuals. The State Board of Health furnished the serum. The University, State Society of Medical Technologists, and State Board of Health all cooperated in sponsoring the effort. The State Board of Health first considered paying a Medical Technologist for doing the work, but instead the Technologists donated their services and will use the money for a Seminar. One of the "bright spots" was accomplished by using 1"x3" glossy finish carboard for typing (saved washing slides). The cardboard was discarded and burned. Notables present included Bob Hope, Mrs. Drummond (who was there for the 4-H career conference), and Dr. Montgomery. A suggestion by Miss Lehman: "We need a Career Exhibit badly. Why can't the American Society do that?" So we see the INDIANA SOCIETY of Medical Technologists has accomplished something out of the ordinary. WHAT HAS YOUR SOCIETY DONE that the rest of us might follow?

*If your state society has participated in some unusual or especially outstanding activities, please let us know. We can make this a regular feature of the AJMT if you are interested and will send in the material.

THE GAVEL

By the time you read this, your first elected representatives as full members of the Board of Registry will have attended their first meeting of that Board, so watch this page in the next Journal for a report.

All your standing committees are completely "staffed" now and are hard at work in your behalf. The importance of cooperating with them in that work cannot be over-stressed; without the help of all our members, our work is ineffective. Besides, these people give a great slice of their "spare" time, a scarce commodity in our profession, and they deserve the best in encouragement and assistance. Tell them of your appreciation, and remember that actions speak louder than words.

Our state society leaders gave us wonderful support when they were asked, in August, to wire their senators in support of the bill authorizing commissioning of women in medical specialties in our Armed Forces. Let me publicly thank them in behalf of us all. With the valuable assistance they gave, your committee representatives on the Civil Service-Armed Service committee have been enabled to inform us that qualified medical technologists may now receive commissions. That is, indeed, a great step forward for our profession. Orchids to our committee and to our state presidents!

Now, as Gabriel Heatter says, a personal word, if you please. As of November 12, your president will be Vernal Johnson Schene, 523½ East Warner, Guthrie, Oklahoma. I hope she will be a satisfactory president; I know she will try.

V. J. S.

IN MEMORIAM
ROY RACHFORD KRACKE
1897 - 1950

Physician, Teacher, Scholar, Author, Pathologist, and Master Medical Technician

The sudden death of Dr. Kracke on the 27th of June has been a terrific shock to his many friends and admirers.

His standard for achievement has come to us through several of his articles which have been published since his untimely death. In each he has made reference to his motto, "second to none." His superlative code of ethics has thus left as a challenge to be met in every field of medical practice.

Those of us who knew him as a teacher will always remember him as an

outstanding scholar who spoke softly but clearly, using exquisite diction, and presenting a mass of factual subject matter with a directness "second to none." The organization of his teaching material was such that his lectures were of value beyond the date of examination.

In final tribute, Dr. Kracke was a physician who lived a daily example of the Hippocratic oath, a teacher who was a *Natural*, an outstanding student and scholar in the field of medical technology, and a pathologist who never requested a technologist to perform a test that he himself could not do as well, if not a great deal better—add to these his accomplishments as an author and his contributions to the fields of medicine and pathology—and we, as medical technologists have an example of a life by which to pattern our future—through art and through scholarship in medical technology, "second to none."

Clara M. Becton

ABSTRACTS

OCCUPATIONAL VIRUS HEPATITUS. By Clifford Kuh and Walter E. Ward, Berkeley, Calif. J.A.M.A., 143: 631 (June) '50.

As a measure of protecting themselves against virus hepatitus, these authors recommend that laboratory technicians handling blood samples should avoid getting blood on the hands. Gloves should be worn when drawing blood from a person with infectious hepatitus. Attention is drawn to the importance of washing hands after exposure to human blood or blood products. These authors report seven cases of virus hepatitus among laboratory personnel who were presumably exposed to a source of infection while at work.

LIMITED USEFULNESS OF MALE AMPHIBIA FOR PREGNANCY TESTS. By O. J. Pollak, Jour. Lab. and Clin. Med. 36: 127 (July) '50.

Attention is called to an observation the author made in a series of 200 pregnancy tests using 2 male frogs and 1 rabbit as the test animals. During the summer months 20% false negative results were obtained. In his opinion negative frog tests must be confirmed by other pregnancy tests, especially during certain seasons of the year.

THE USE OF PROTEIN SILVER FOR STAINING PROTOZOA. By Norman Moskowitz, Stain Technol. 25: 17 (Jan.) '50.

Although the material used in these studies was primarily from rodents, it is the opinion of the abstractor that the procedures could be applied to human material in cases which needed better differentiation for classification.

1. Fix smears in Holland's cupric picro-formal mixture (See Kirby J. Parasit. 2. 31: 163, 1945).
2. Wash in 50% alcohol to remove fixative.
3. Harden smears in 70% alcohol.
4. Hydrate smears.
5. Bleach.
   a. 0.5% potassium permangenate 2 min.
   b. Wash twice in distilled water.
   c. 5% oxalic acid for two minutes.
   d. Wash three times in distilled water.
6. Impregnate smears in protein silver solution 36 hours at 35° C.
   0.9 gm. gelatin (Knox or Eimer & Amend, USP) is dissolved in 100 ml. distilled H_2O by aid of heat. Add 1.0 gm. Silver Nitrate at 60° C., 1-2 drops of M/10 NaOH to 10 ml. sol. enhances impregnation and staining.
7. Reduction of silver.
   Rinse impregnated smears in distilled water and treat for 5 min. in a reducing bath consisting of 1 gm. hydro quinone and 5 gm. Sodium Sulfite per 100 ml. distilled H_2O.

8. Wash in distilled water 1 minute.
9. Toning—Immerse preprations for 4 minutes in 0.2% yellow gold chloride.
10. Rinse in distilled water.
11. Treat 2% oxalic acid 3 minutes.
12. Wash 1 minute in 3 changes distilled water.
13. 5% Sodium Thiosulphate 7-8 minutes.
14. Wash in running tap water ½ hour.
15. Dehydrate.
16. Mount.

BOOK REVIEWS

RECENT ADVANCES IN CHEMOTHERAPY: Third Edition. Vol. I. By G. M. Findley, C.B.E., Sc. D., M.D., F.R.C.P., Editor, Abstracts of World Surgery, Gynecology, and Obstetrics, British Medical Association, London. 625 Pages; June 14, 1950. The Blakiston Company, Philadelphia and Toronto. $7.50.

This work brings up to date the accepted chemical therapeutic agents indicated in various parasitic infestations. It is a book for physicians and one with which all general practitioners should become acquainted.

MEDICAL PARASITOLOGY. By William G. Sawitz, M.D., Associate Professor of Parasitology Associate in Medicine, The Jefferson College of Philadelphia; Special Consultant, U. S. Public Health Service, Communicable Disease Center, Atlanta, Georgia. 90 illustrations; 296 pages; June 14, 1950. The Blakiston Company, Philadelphia & Toronto. $4.25.

There are a number of interesting features about this new volume. It is not a text but a manual whose function is to serve as a lecture and laboratory guide for students taking a course in medical parasitology. The author has listed the textbooks and journals important in parasitology. Parasitology is outlined—only the most pertinent facts are given. There is a chapter on technics as well as one on treatment. Teachers of medical technologists will find excellent material in this small volume.

This book does not compare with the larger and more complete works; nevertheless, it should prove a popular edition because of the cost and of the forthright manner in which the material is presented.

HELPFUL HINTS TO THE DIABETIC: By William S. Collens, M.D. Chief of the Diabetic Clinic, Maimonides Hospital, Brooklyn, and Louis C. Boas, Chief of the Diabetic Clinic, Green Point Hospital. 135 pages, 64 illustrations, 21 figures. Price $3.50.

This little manual has been prepared expressly for diabetic patients. Your reviewer was so impressed with the volume that she requested one for special review.

The material presented is, in the opinion of the authors, what a diabetic should know about diabetes, its treatment, and how it is managed. The chapter on *The Examination of Urine* is par excellent, and should be read by every person teaching a patient how to examine the urine for sugar. Moreover, those medical technologists who do not have a college background in the medical sciences will find a wealth of useful information in this book.

With appropriate permission from physicians in charge of laboratories it should be possible to place this timely little book in the hands of those diabetics who desire a more thorough understanding of the disease.

Every person handling diabetics will find a lot of very useful information in this little book. Interns, physicians and medical technologists should read it because it contains those essentials necessary for adequate control of the disease. The style is very readable. Less than two hours were needed by the reviewer to read the volume through carefully. Every student medical technologist should be required to read this book in that it gives a terse summary of the problems of the diabetic patient.

COAGULATION, THROMBOSIS, AND DICUMAROL. With an Appendix on Related Laboratory Procedures, by Shepard Shapiro, M. D., *Assistant Professor of Clinical Medicine, New York University College of Medicine; Visiting Physician, Third (New York University) Medical Division, Goldwater Memorial Hospital; Associate Physician, Lincoln Hospital;* and Murray Weiner, B.S., M.S., M.D., *Fellow in Medicine, New York University College of Medicine; Research Assistant, Third (New York University) Medical Division, Goldwater Memorial Hospital; Assistant Visiting Physician, Willard Parker Hospital Chest Service; Clinical Assistant Visiting Physician, Bellevue Hospital.* 131 pages. Brooklyn Medical Press, New York, N. Y., publishers, 1950. Price $5.50.

This little volume has been written primarily for physicians. Nevertheless, the tests described in the appendix will be of great practical importance to medical technologists if these methods are accepted by the medical profession, particularly clinical pathologists and clinicians.

Although this volume is quite small, it gives a synopsis of current theoretical and practical aspects of present day anticoagulant therapy as well as setting forth a practical approach to the control of thrombosis and hemorrhagic conditions into one book.

A good summary of the factual knowledge, which in the hands of the authors has proved to be dependable and clinically valuable is presented to the busy practitioner, clinical pathologist and medical technologist.

THE DIAGNOSIS OF SALMONELLA TYPES: By F. Kauffmann, M.D. Chief, International Salmonella Center, State Serum Institute, Copenhagen, Denmark. 86 pages. Springfield, Ill., 1950. Charles C. Thomas, Publisher. Price $2.25.

This timely little monograph is the 62nd publication in the American Lecture Series published by Mr. Thomas. The classification of the Salmonella group has almost become a specialty in bacteriology. Dr Kauffmann needs no introduction to bacteriologists. Therefore, it is a delightful fact that we now have a book which brings into a few pages the accepted factual knowledge of the somewhat baffling subject.

Laboratories doing large volumes of work will find this new volume a must. Technics and methods are given.

HISTOLOGY: By Arthur Worth Ham, M. B. Professor of Anatomy, in charge of Histology, in the Faculties of Medicine and Dentistry, University of Toronto. 756 pages, 445 figures. Philadelphia, 1950. J. B. Lippincott Company. Price $10.00.

This volume on histology is, in the opinion of the reviewer, the most readable among the current books on this subject. It is a beginner's text. The importance of understanding physiology in terms of histology cannot be over-emphasized. The author is truly a versatile histologist, if he were not, it would be utterly impossible to have produced a volume that builds bit by bit, the essential information needed in understanding this complex subject.

Of particular interest to medical technologists are the chapters on the Blood Cells and Hematopoietic Tissue. There are not many books which give the functions of these elements in such a direct manner. The living tissue, rather than a prepared histologic slide, seems to be constantly before the reader.

Beginning medical technologists in surgical pathology should read part I in this book. This is an introduction of the methods used in preparing histological material.

Another fact that must be considered at this point is that Dr. Ham teaches those medical students who have the privilege of studying pathology under the one and only Dr. William Boyd. In order to understand pathology, histology is a must, and it is to this end that this amazing volume has been prepared and written.

All medical libraries should have this book on the shelves as well as hospitals and laboratories. To those medical technologists desiring more information about histology and blood cells, this book will fill a void.

ANNOUNCEMENTS

CIVIL SERVICE-ARMED SERVICES COMMITTEE ARMED SERVICES RATINGS

A message from your Civil Service and Armed Services Committee: H. R. 4384 (the bill we supported, remember?) has passed the Senate and House and is now in conference as there are reconciliations necessary. Qualified Medical Technologists (both men and women) ARE being commissioned in the Reserve Medical Corps of all branches of the Armed Services. This was announced in a directive dated August 21, 1950.

Your Committee is of the opinion that this favorable response to our resolution (requesting commissions) was greatly hastened by the excellent cooperation shown by the State Presidents and various individual members to the request from Miss Johnson to the states to wire their Senators concerning H.R. 4384 and to write follow-up letters explaining who and what medical technologists are and do. This is an excellent example of what can be done nationally through our state societies in support of a common cause. For example, Miss Johnson called Miss Matthaei Saturday A. M., August 12, dictating a letter which went to the State Presidents—by 3 P.M. of the same day the letters were in the mail and by Wednesday, August 16, your Committee Chairman was receiving copies of letters sent by Senators to the Presidents in their States assuring them of those Senators' support. It has strengthened our cause and the same result has been achieved by directive sent to the Recruiting divisions of all the Armed Services.

STATUS OF CIVIL SERVICE RATINGS

Previous to the passage of the Classification Act of 1949 (Public Law 429) all medical technologists' positions were in the subprofessional (SP Series) Services. At the present time the Medical Technician Series (SP 636-O) has not been changed to conform with the provisions of the Classification Act. This Act abolishes the professional, subprofessional, and all other classifications and puts all Civil Service jobs under the so-called General Schedule (GS) grades. Some of the former SP grades were abolished. Some were combined to make other grades (e. g., there are no grades GS-6, GS-8, or GS-10).

At the present time Civil Service recognizes graduates of the A.M.A. Approved Schools for Medical Technology and allows such qualified persons to enter the service as a GS-5 which is the lowest grade of the former Professional Service (P-1). Nurses, Physical Therapists, and other auxiliary medical personnel enter Federal Service at the same grade level and may be classified as high as a GS-12 (old P-5). The highest grades given to Medical Technologists as such, is GS-7 (old SP-8). Therefore, our present request is for two series, one for Medical Technicians—(Laboratory Aides?) with GS-2 through 5, and Medical Technologists Series, GS-5 through 11.

The Classification Section and the Standards and Studies Division of the U. S. Civil Service Commission have informed our joint Committee that when we have submitted all material which they have suggested might be pertinent and that which we think should be included in support of our request, they will review all this material and give an opinion on the feasibility of our proposals. This report will be continued as soon as there are further developments.

Mrs. Lucille Wallace, Chairman

Editor's Note: We of the ASMT owe to Mrs. Lucille Wallace and Miss Mary Eichman a vote of appreciation for their generous and tireless work on the Civil Service-Armed Services Committee. These two members of our society spent many hours during the convention, have studied require-

ments, and have conferred with those in authority, with the result that the medical technologist can now realize that his wish that "they" would do something about Civil Service and military ratings has begun to crystallize into fact. We can also appreciate the efforts of Dr. Frank Queen and other members of the Board of Registry whose advice has been invaluable in this respect. We had hoped to have a complete list of the requirements for military ratings for publication in this issue of the JOURNAL, but the expected report has not reached us from Washington.

PENNSYLVANIA HAS DONE IT!

You can too!

Yes, the Pennsylvania Society has done something about UNESCO. Please take this matter up at your next Board meeting. A recent communication has informed us that used reference books, in good condition, less than ten years old will be welcome. The CARE Book Program is a handy method, and for any other contributions you may contact me, and I will combine our funds to send to the UNESCO office where they will be used as needed. The Pennsylvania Society? Oh yes, they took the lead —they have already sent $10.00 to the CARE Book Program. It's a start, and a good one.

Doris E. Boone, MT (ASCP)
1588 Ansel Road
Cleveland, Ohio.

PRELIMINARY ANNOUNCEMENT OF THE CLEVELAND MEETING OF THE AAAS DECEMBER 26-30, 1950

The 117th meeting of the American Association for the Advancement of Science, the annual meeting for the year 1950, will be a full-scale meeting—with programs in every principal field of science from astronomy and botany to, and including, zoology. All 17 of the Association's sections and subsections, and more than 40 participating societies and organizations, are completing plans for an aggregate of more than 200 sessions.

There will be a considerable number of outstanding symposia. The list of special sessions includes the AAAS Presidential Address, and addresses sponsored by the Academy Conference, the National Geographic Society, the Scientific Research Society of America, the Society of the Sigma Xi, and the United Chapters of Phi Beta Kappa.

An extensive series of tours to museums, laboratories, and industrial plants of the Cleveland area has been planned. Some of these will be of particular interest to chemists and are being included as a part of the program of Section C.

The hotels of Cleveland have pledged themselves to provide ample housing for those attending the 117th meeting. Housing will be taken care of by the experienced personnel of the Cleveland Convention Bureau, with hotels confirming reservations directly. Announcements and and coupons for reservations will appear in *Science* and *The Scientific Month-*

ly, beginning the last of August. The hotels include the Statler (headquarters of the AAAS,) the Hollenden, the Carter, the Allerton, the Olmsted, and the Auditorium Hotel—the last particularly convenient for exhibitors.

At the New York Meeting, the Council of the Association voted overwhelmingly to continue with advance registration. Plans are already made to ensure the mailing of the General Program to advance registrants not later than December 1 (as compared with December 6-9 last year), *by first class mail.*

RAYMOND L. TAYLOR,
Assistant Administrative Secretary
AAAS
1515 Massachusetts Avenue, N. W.
Washington, D. C.

PLEASE NOTE THE FOLLOWING AVAILABLE AWARDS FOR AMERICAN SOCIETY OF MEDICAL TECHNOLOGY MEMBERS

1. Awards from Board of Registry for Scientific Papers and Scientific Exhibits.
2. ASMT Convention Awards for Scientific Papers and Scientific Exhibits.
3. The Hillkowitz Memorial Contest.
4. Parasitology: Best paper on parasitology—$25 award.

This should be an inspiration to all Medical Technologists to write a Scientific Paper or plan an Exhibit.

Please Note the Following Propram Rules:

1. The deadline date for papers to be received by the Program Committee from individuals desiring to present papers in Swampscott and likewise to compete for the Convention Awards is March 15, 1951.
2. Only ASMT members are eligible to compete for Convention Awards. All competitive papers must be presented in persen or by proxy at convention time.
3. All Papers read or submitted to the Society become the property of ASMT and may be published in the *American Journal of Medical Technology.*
4. The time limit for reading the paper on the Program is 15 to 20 minutes exclusive of showing slides—the remainder of approximately 30 minutes is to be given to discussion. Papers may be longer and abstracted for Program purposes.
5. All audio visual aids and professional technicians to operate them will be supplied by the Speakers Supplies Committee. Standard lanterns (3½ by 4) will be furnished unless otherwise requested. Please indicate what aids are needed.
6. **Five** (5) copies of your manuscript must be submitted to the Program Committee Chairman. These must be typewritten, double spaced on regular size typewriter paper.
7. **Two** (2) copies of the manuscript must be submitted by all those NOT COMPETING for ASMT AWARDS and subject to the above instructions.
8. **Prize Papers** from State Contests to be considered for presentation and further awards, must be in the hands of the Program Committee Chairman by March 15, 1951.

Program Committee:
Helen J. Madden, Chairman, 86 Jersey St., Boston 15, Mass.
Sr. Mary James, Santa Rosa Hospital, San Antonio, Texas.
Sr. M. Alcuin, OSB, College of St. Scholastica, Duluth 2, Minnesota.
Marie Colburn, 2046 Mt. Washington, Colorado Springs, Colorado.
Ellen Skirmont, 5493 So. Cornell Ave., Chicago, Illinois.
Lydia Brownhill, Meridan Hospital, Meridan, Connecticut.
Ruth Cleo Ball, 3000 Polk Ave., Ogden, Utah.

NOMINATIONS AND ELECTIONS

The Nominations and Elections Committee wishes to call attention to the list of officers to be elected at the next annual meeting of the American Society of Medical Technologists. The committee has notified all officers and affiliated societies of the ASMT by Newsletter to send in their selections for these offices. The members of the committee would also appreciate any suggestions from the individual members of the society. NOW is the time to voice your opinions as to whom you would like to have nominated to these positions. ALL SUGGESTIONS MUST BE IN THE HANDS OF THE COMMITTEE MEMBERS BY DECEMBER 1, 1950.

The offices to be filled and those now holding these offices are as follows:

President-elect: Lavina White.

Recording Secretary: Sister Eugene Marie (not eligible for re-election).

Treasurer: (three year term) Loretta Laughlin (not eligible for re-election).

Board of Directors: (three year term) Mary Nix and Louise Vance.

It is important that we have the names and qualifications of the proposed candidates from those who best know their qualifications. They must be members of the ASMT in good standing and must have been members for at least two years.

Please send communications at once to:

Violetta A. Wakefield, 1209 N. 34th St., Fort Smith, Ark. (chairman)
Louise Miller, 2813 Elizabeth Road, Hampton, Va.
Hazel Current, 918 17th St., Santa Monica, Calif.
Oscar M. Alton, 2036 Tyler Lane, Louisville, Ky.
Eleanor Fulton, 506 Amadore Apts., Saginaw, Mich.
Lucile Harris, Hendrick Memorial Hospital, Abilene, Texas.

FIRST SCHOLARSHIP FOR MEDICAL TECHNOLOGY OFFERED

Alpha Mu Tau Fraternity marks the mid-century with an eventful announcement for the profession of Medical Technology. The announcement of the First Scholarship for Medical Technology of $100.00 was made public during the annual convention of the American Society of Medical Technologists in Houston, Texas, June 11-15, 1950. This scholarship was offered to any qualifying senior college student of medical technology in the United States. Applicants may obtain full information from the Scholarship Chairman, Miss Rachel Lehman, M.T. (ASCP), Indiana University School of Medicine, Indianapolis, Indiana.

Alpha Mu Tau Fraternity is a professional organization incorporated in the State of Illinois in February, 1948, with its objective the advancement of Medical Technology as a profession, so that it can and will be recognized by other scientific groups as a necessary and integral part of the medical profession. A primary means to gain this objective is to grant scholarships to worthy students, who do not have funds immediately available to pursue higher education in this field. Membership is limited to Medical Technologists, who are in good standing with ASMT and who have made an outstanding contribution to Medical Technology. Meetings of this Fraternity are held annually, at the place and time of the National Convention of the American Society of Medical Technologists.

The following officers were elected for the fiscal year 1950-1951:

PRESIDENT: Miss Mary Nix, 4931 W. E. Gilsen, Apt. 202, Portland, Oregon.
VICE-PRESIDENT: Miss Ellen Skirmont, 5493 S. Cornell Ave., Chicago 15, Ill.
SECRETARY: Mrs. Winogene McIntyre, St. John, Washington.
TREASURER: Miss Faith Dravis, P. O. Box 133, Prescott, Wisconsin.
BOARD OF DIRECTORS: Miss Rachel Lehman, 3939 North Capitol, Indianapolis, Indiana.

From the Illinois Journal of Medical Technology, Vol. 2, No. 1.

FOR 1951 CONVENTION SEALS! write to Dorothy Prest, 78 School St., Manchester, Mass. 1c each up to 400, $4.00 for 500, $7.50 for 1000.

STANDING COMMITTEES 1950-51

Public Relations

Chairman:	Frieda Claussen, Minnesota	2 years
	Mary Nix, Oregon	2 years
	Lavina White, Colorado	1 year
	Doris Boon, Ohio	1 year
	Josephine Pyle, Connecticut	3 years
	Marian Baker, Pennsylvania	3 years

Constitution and By-Laws

Chairman:	Margaret Haraway, Oklahoma	3 years
	Allyne Lawless, Colorado	1 year
	Sister M. Veronica, South Dakota	1 year
	Ada Gregory Slior, Kansas	2 years
	Mary Yeaton, Massachusetts	2 years
	John Mooty, Wisconsin	3 years

Nominations and Elections

Chairman:	Violetta Wakefield, Arkansas	2 years
	Hazel Current, California	1 year
	Oscar M. Alton, Kentucky	1 year
	Louise Miller, Virginia	2 years
	Eleanor Fulton, Michigan	3 years
	Lucile Harris, Texas	3 years

Research

Chairman:	Hazel Suessenguth, Ohio	3 years
	Hartzell Payne, Indiana	1 year
	Elvyn Scott, Delaware	2 years
	Ruth Guy, California	2 years
	Margret Hughes, Oregon	3 years
	Dorothy Hitchcock, Michigan	1 year

Membership

Chairman:	Ida Reilly, Virginia	3 years
	Mrs. Frances Moore Kenworthy, Illinois	3 years
	Mrs. Lucille Wallace, Minnesota	2 years
	Eleanor Brenny, Florida	2 years
	Sr. Chas. Miriam, New Mexico	1 year
	Winogene McIntyre, Washington	1 year

Finance

Chairman:	Mary Eichman, Pennsylvania	2 years
	Louise Vance, Illinois	2 years
	Oscar Stewart, Oklahoma	1 year
	John Hannon, Ohio	1 year
	Loretta Laughlin, Minnesota	3 years
	Kathryn Dean, Maryand	3 years

Legislation

Chairman:	Sadie Cartwright, Georgia	3 years
	Wilbert Zimmer, Ohio	1 year
	Grace Mary Ederer, Minnesota	1 year
	Beatrice Allison, New York	2 years
	Barbara Isbell, California	2 years
	Wade Marsh, Tennessee	3 years

Education

Chairman:	Rose Hackman, Colorado	2 years
	Estelle Downer, Wisconsin (Seminar Fund)	1 year
	Mary Frances James, Alabama	1 year
	Ruth Feucht, Illinois	2 years
	Joy Holm, Louisiana	3 years
	Elizabeth Frey, New York	3 years

Standards and Studies

Chairman:	Sister M. Dolorosa (Pope), Missouri	1 year
	Elinor Judd, Massachusetts	1 year
	Carolyn Martin, Georgia	2 years
	Agnes Marie Lyman, Oregon	2 years
	Anne Priest, North Carolina	3 years
	Sister Leo Rita, Missouri	3 years

OPPORTUNITIES FOR UNIVERSITY LECTURING AND ADVANCED RESEARCH

Approximately 300 Fulbright awards are available to U. S. citizens for university lecturing and advanced research abroad for the acamedic year 1951-52 as announced by the Department of State on June 5, 1950. Many of the lecturing opportunities are in specific subjects at designated universities. Others are unspecified as to subject and institution and applications for lecturing will, therefore, be accepted in subjects other than those listed.

The closing date for applications was October 15, 1950. Now is the time to begin thinking about your plans for 1952-53.

Many of the awards are open to applicants in medicine and the various medical specialties, public health, nursing, dentistry and the basic sciences. Countries now participating in the program are Australia, Belgium-Luxembourg (including the Belgian Congo), Burma, Egypt, France, Greece, India, Iran, Italy, Netherlands, New Zealand, Norway, the Philippines, Turkey, and the United Kingdom, including the British Colonial Dependencies.

The Fulbright Act authorizes the Department of State to use certain currencies and credits acquired through the sale of surplus property abroad for programs of educational exchange with other nations. Awards are ordinarily made for one academic year, although in exceptional cases applications will be considered for periods of not less than six months. Grants for teaching or research usually include round-trip transportation for the grantee, a maintenance stipend, including certain allowances for dependents, and a small supplemental allowance for travel and equipment purchasable broad if necessary.

Requests for application forms and for further information concerning opportunities for visiting lecturers, research scholars and specialists should be addressed to the Committee on International Exchange of Persons, Conference Board of Associated Research Councils, 2101 Constitution Avenue, Washington 25, D. C.

DUES!

By this time you have paid your 1950-51 dues or you have received a "Second Notice" as a reminder that you want to stop now and write a check payable to your state treasurer. If for some reason, you won't be able to pay dues this year, please send some notice of that fact (a penny post card will do) to your treasurer or to the Executive Office of ASMT, so that when you meet better times, you'll be able to rejoin your national organization without penalty. We don't want to lose any members but we would rather file your cards away as "resigned in good standing" than just write "Dropped for non-payment of dues." This will be the last issue of the journal mailed to those who do not get their dues paid by Jan. 15, 1951. We would appreciate your notifying us *promptly* of any change in your name or address. If you are not receiving your journals regularly, it may be due to the fact that you didn't let us know your new address. After each journal mailing, we find ourselves paying an excessive amount of "postage due." Many post offices don't return the journals to us, but send a "postage due" card with notice that the addressee has moved. Frequently these address changes are not legible. It would be much better if *you* let us know *immediately* you are located at a new address. So PLEASE:

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